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A STUDY OF THE CHEMICAL AND PHYSICAL PROPERTIES OF REMADE MILK¹

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The rapid increase in the development of the powdered milk industry during recent years and the increasing consumption of this product in liquid form has raised several important questions of interest both to the manufacturer of the powdered milk, and to the consumer who buys it, either in reconstituted or reconstructed form, possibly as "fresh" milk. The dairy control laboratories of our large cities whose duty it is to protect the milk supply of their respective communities are also interested in the advent of powdered milk for use in the standardization of market milk or its sale as whole milk after reconstitution from whole milk powder, or its reconstruction from skim milk powder and butter.

Among the important questions raised in this new field of the dairy industry is the question whether powdered milk exhibits the same chemical and physical properties after remaking it into fluid milk as did the original milk from which the powder was produced. Other important questions have to do with the nutritional, especially the vitamine, efficiency of remade milk, and also its safety from a bacteriological point of view.

It is not the purpose of this paper to present data on the latter two aspects of the problem. It is desired, however, to report a study of certain of the chemical and physical properties of milk either (1) reconstituted from whole or partially skimmed

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milk powders or (2) reconstructed from skim milk powder and butter. In undertaking this study it was not our purpose to try to find a test for remade milk, but merely to examine its properties from a general point of view. In so doing, it was recognized at the outset that there are essentially only three types of powdered milk on the market at the present time. One type has been dried at atmospheric pressure in a thin film on a slowly revolving heated drum according to the basic patents now known as the Just-Hatmaker process. Another, more recent type, has been dried by spraying the previously condensed milk under hydraulic pressure into an essentially closed chamber through which a stream of heated air is continually passing. This process is exemplified by the milk manufactured by the Merrill-Soule process. A third type differs from the Merrill-Soule process in that the milk which may or may not be previously condensed, is sprayed by centrifugal force into a tower through which heated air is being circulated in a cyclonic or some similar manner. Patents controlled by the California Central Creameries Company of Los Angeles, California, and the International Dry Milk Company of Minneapolis, Minnesota, illustrate this type of process.

EXPERIMENTAL

The experiments reported below include an examination of milk remade from each of the three types of process of manufacture and also of milk reconstructed from skim milk powder, water, and sweet, unsalted butter. These milks will hereafter be referred to as drum process, pressure spray process, centrifugal spray process and reconstructed milks, respectively. Fresh powder was obtained at the beginning of the study. From time to time fresh samples made by the centrifugal spray process were obtained. The stock of pressure spray powder was not replenished but new cans from the original lot were opened from time to time. The same lot of skim milk powder was used throughout the experiments in making the reconstructed milk. The drum process powder was renewed once during the course of the study.

Method of remaking the milk. An ordinary egg beater and Dazey churn were used in remaking the milks from whole milk or partly skimmed milk powder and water. Usually just enough milk was made up at one time for the experiment to be conducted. Tap water was used at a temperature of 60–65°C. except in the experiments on freezing point, specific conductivity and buffer value, where distilled water was used. The samples were allowed to cool before using.

A De Laval emulsor no. 2 was used in reconstructing the milk from skim milk powder, sweet butter and water, following the directions put out by the manufacturers of the machine.

PHYSICAL PROPERTIES OF REMADE MILK

Freezing point. The freezing point is one of the least variable of the physical properties of milk. It has been long recognized as the most sensitive test available for the detection of added water in milk, as has been recently confirmed anew by Hortvet (1).

The purpose of examining this property of remade milk was to determine how closely the reconstituted and reconstructed products resemble fresh milk when made up according to the directions given by the manufacturers. In case divergencies occurred it was planned to determine, if possible, the correct proportions of powder and water to use to obtain a product showing the normal freezing point of fresh milk. It was believed that such data would be of value to those making reconstituted or reconstructed milk as an aid in obtaining a product which would resemble most closely fresh cow's milk. It was planned, also, to use the data thus secured in our own subsequent experiments in remaking the milk for the examination of other properties.

Evenson (2) has recently determined the freezing point of milk remade from various kinds of powder. The exact proportions used by him in reforming the fluid milk are not stated. Some variations from the accepted average normal of -0.55°C . were found, but the conclusion drawn was that the differences

are not great enough to make the freezing point of value in detecting mixtures of natural and remade milk, which Evenson was seeking to do.

In our experiments the pressure and centrifugal spray whole milk powders were made up in the proportion of 1 part by weight of powder to 7 parts by weight of water, yielding milk with approximately 12.5 per cent total solids. In the case of the drum process powder, made from partly skimmed milk, the directions of the manufacturer call for mixing 1 part by weight of powder and 8 parts by weight of water, yielding a product containing approximately 11 per cent total solids. In the case of the reconstructed milk 2.29 pounds of skim milk powder were dissolved in 21.67 pounds of water and emulsified with 1.04 pounds of sweet cream butter, forming 25 pounds of milk testing 3.5 per cent fat.

The freezing point was determined by means of the newly designed Hortvet (1) cryoscope, the thermometer having previously been standardized, using solutions of pure sucrose.

The results of these experiments are given in table 1, which shows the freezing point depression as obtained by using the proportions of ingredients recommended by the manufacturers and also the proportions which it was found necessary to employ to secure a normal freezing point.

The data in table 1 show that the manufacturers of the drum process, partly skimmed milk powder, recommend slightly more powder than necessary to give a milk showing a normal freezing point, while the manufacturers of the centrifugal spray process powder recommend slightly less powder than is needed to secure a normal result. It is possible that the latter result is due to the fact that the heat employed in the process denatures slightly the hydrophylic colloids of the milk so that they do not bind back the water which they naturally contained, the effect being that the milk has been slightly watered. Other factors which may be involved are (1) a high moisture content of the powder, (2) a lower total solid content of the original milk than the standard adopted by the manufacturer.

Specific gravity. The specific gravity of the various kinds of remade milk was determined at 60°F. two hours after being made up according to the proportions giving a normal freezing point. The figures obtained were as follows: drum process milk, 1.0336; pressure spray milk, 1.032; centrifugal spray milk, 1.0314; re-constructed milk, 1.0314. The figures are seen to lie within the limits of variation of normal milk.

Viscosity. It is generally recognized at the present time that the viscosity or fluidity of milk is determined largely by the constituents of milk which are in colloidal solution, namely, the calcium caseinate, the albumin and globulin, and the colloidal

TABLE 1
Freezing point of remade milk

KIND OF MILK	PROPORTIONS RECOMMENDED BY MANUFACTURERS			Δ °C.	PROPORTIONS FOUND NECESSARY FOR NORMAL			Δ °C.
	Powder	Water	Butter, 84 per cent fat		Powder	Water	Butter, 84 per cent fat	
	per cent	per cent	per cent		per cent	per cent	per cent	
Normal cow's milk.....				-0.555				-0.555
Drum.....	11.1	88.9		-0.655	10.6	89.4		-0.550
Pressure spray.....	12.5	87.5		-0.545	12.5	87.5		-0.545
Centrifugal spray.....	12.5	87.5		-0.500	12.8	87.2		-0.548
Reconstructed.....	9.16	86.68	4.16	-0.560	9.16	86.7	4.16	-0.560

di-calcium phosphate, rather than by the fat which is present in particles whose size is considerably above the colloidal realm. Any process of powdering milk which has a profound effect on the colloids of the milk would therefore be expected to manifest itself in the viscosity or fluidity of the remade product.

In determining this property the milks were remade in the proportions giving a normal freezing point and the observations made at 25°C., while the milk was fresh, and again after standing for 24 hours at 7°C., the reading again being taken at 25°C.

A McMichael viscosimeter was used but the calibrated disc was rotated as in the Doolittle (3) or Mojonnier method. The average of four readings was calculated in terms of centipoises

or absolute units using tables which had been worked out for the instrument in this laboratory.

The results are shown in table 2, in comparison with readings made on freshly pasteurized milk. The data indicate a slight alteration of the colloids in all cases. This is least noticeable in the case of the pressure spray milk and most evident for the drum process milk. The decline in viscosity on standing in the case of the drum process milk is to be explained on the grounds of a greater dispersion of the colloidal material on standing. The increase occurring in all the other cases, including the natural milk, is presumably a hydration effect in the realm of greatest colloidalilty.

TABLE 2
Viscosity of remade milk

KIND OF MILK	TEMPERATURE	VISCOSITY WHEN FRESH	VISCOSITY AFTER 24 HOURS AT 7°C.
	°C.	centipoise	centipoise
Pasteurized.....	25	1.39	3.25
Drum process.....	25	7.36	5.91
Pressure spray.....	25	1.79	3.64
Centrifugal spray.....	25	2.25	4.10
Reconstructed.....	25	1.94	2.29

Specific conductivity. The specific conductivity was determined in a Freas cell at 25°C. The constant of this cell was found to be 0.34202. The data obtained are given in table 3. With the exception of the drum process milk practically the same values were obtained for all the milks, including the normal milk samples used as a check. The high conductivity of the drum process milk may be due in part to the partial skimming, Durand and Stevenson (4) having shown the skim milk has a higher conductivity than whole milk.

Creaming ability. The amount of cream which rises on milk is used by the public to judge the richness of the milk, notwithstanding the fact that this is frequently an erroneous standard. Remade milk for the most part shows little or no rise of fat to form a cream layer. In the case of milk remade from powder

manufactured by the pressure spray process no cream rises at all because the fat is thoroughly homogenized in powdering the milk. The milk remade from the drum process powder gave a small cream line on standing, the volume of cream amounting to about 2.5 per cent. It will be recalled that this powder is made from partly skimmed milk. Milk which was reconstructed from skim milk powder and butter also threw up a small cream layer (about 3 per cent), but the cream was very rich, almost like butter. It consisted of large globules very closely packed together. Only in the case of the centrifugal spray powder was anything like a normal cream line obtained. This milk, after remaking, showed a cream layer of about 13 per cent, which contrasts very favorably with the usual cream layer of 14 to

TABLE 3
Specific conductivity of remade milk

KIND OF MILK	NUMBER OF DETERMINATIONS	AVERAGE SPECIFIC CONDUCTIVITY
		<i>mhos</i>
Fresh, pasteurized.....	4	0.00547
Drum process.....	8	0.00625
Pressure spray.....	8	0.00545
Centrifugal spray.....	12	0.00560
Reconstructed.....	8	0.00570

16 per cent which is obtained for the average pasteurized market milk. The milk in all these experiments, was allowed to cream at 10°C.

General physical appearance on standing. In addition to the cream layer other features of the remade milks were noted. The milks remade from the pressure spray powder and reconstructed from pressure spray skim milk powder and butter exhibited no abnormalities. The milk remade from the centrifugal spray powder, however, showed a large volume of foam-like material on the surface which consisted of floating, partly dissolved granules of powder. The dispersed fat in this milk was very prone to churn when the milk was shaken. It was, in fact, found necessary to exert care in remaking the milk to avoid this churning of the fat.

In the case of the milk made from the drum process powder there was always a separation of solids-not-fat after standing over night. The fluid at this time exhibited 3 layers, a layer of casein-like material at the bottom, a layer of clear serum above this, and a layer of cream on the surface.

CHEMICAL PROPERTIES OF REMADE MILK

Soluble proteins. The so-called soluble proteins of milk, the albumin and globulin, which in reality are present in a colloidal state, begin to coagulate at 72°C. Although even long continued boiling fails to remove all of these two proteins from milk, as one of us (5) has recently shown, nevertheless the content of soluble protein in remade milks should give roughly an index of the maximum temperature employed during the drying process. The analysis of remade milks for total soluble protein should at least indicate whether the powder ever attained a temperature of 72°C. during the process.

The method employed in our experiments was to remove the casein from 10 cc. portions of the remade milk by precipitation with acetic acid, following the usual A. O. A. C. method. The casein was washed, however, with acetic acid water adjusted to a pH of 4.8. The filtrate and washings were neutralized to a faint pink with NaOH solution, using phenolphthalein as indicator and the soluble proteins precipitated with 50 cc. of Almen's tannic acid reagent. After standing for one hour the precipitate was filtered off, washed, and the nitrogen determined in the precipitate by the usual Kjeldahl method.

The results obtained are shown in table 4. The data show that the soluble proteins have been materially lessened during each of the processes except the centrifugal spray, indicating that the heat in these cases must have reached 72°C. The drum process powder obviously attains the highest temperature and the centrifugal spray powder the lowest. The partial coagulation of the soluble proteins in the case of the pressure spray powder probably occurs either during the preliminary condensing process or while the powder is subjected to the continued blast of hot air as it lies in the powdering chamber during a "run."

Rennet coagulability. The possibility of using remade milk for cheese manufacture is worthy of consideration. To be useful for this purpose, however, the remade milk should at least possess the rennet coagulability of pasteurized milk.

In our tests the Marshall rennet cup was used to determine the speed of coagulation and the character of the curd. Several tests were run on each milk with uniform results. Milk made from the centrifugal spray powder was found to resemble closely pasteurized milk both as to the speed of coagulation and character of clot. The pressure spray milk ranked next, a little more time being required to stop the flow of milk from the bottom of the Marshall cup, and the curd being less firm. The drum process

TABLE 4
Soluble protein content of remade milk

KIND OF MILK	AMOUNT N OBTAINED	SOLUBLE PROTEIN (N \times 6.38)
	mgm. per 10 cc.	per cent
Normal.....	10 to 12	0.64 to 0.77
Centrifugal spray.....	11.05	0.70
Pressure spray.....	8.82	0.56
Reconstructed.....	8.17	0.52
Drum.....	7.22	0.46

milk and the reconstructed milk failed to clot, a very fine coagulation of the casein occurring instead.

The failure of the drum process milk to clot normally is in line with the observations of others (2). The reason for the failure of the reconstructed milk to clot probably lies in the repasteurization of the milk during the process of remaking. This point, however, needs further investigation.

HYDROGEN ION CONCENTRATION AND BUFFER VALUE

Evenson (2), has determined the hydrogen ion concentration of remade milks and also their buffer value as compared with pasteurized milk. No differences were noted by him except in a case where the milk had obviously been treated with alkali. He has not reported his results in detail.

We also performed essentially the same experiments with the types of remade milk under investigation in comparison with freshly pasteurized milk. In determining the buffer value of the milks towards both acid and alkali 25 cc. portions of the milk were treated with increasing amounts of 0.1 N lactic acid and lime water, respectively, and the hydrogen ion concentration of the mixture determined electrometrically. The data are presented in table 5. The portion of these data near the initial value are shown graphically in chart 1, after calculating the concentration of 0.1N acid and alkali, respectively, in the samples tested for hydrogen ion concentration.

TABLE 5
Buffer value of remade milk

AMOUNT OF ACID AND ALKALI ADDED TO 25 CC.	PASTEURIZED		DRUM		PRESSURE SPRAY		CENTRIFUGAL SPRAY		RECONSTRUCTED	
	Acid	Alkali	Acid	Alkali	Acid	Alkali	Acid	Alkali	Acid	Alkali
cc.	pH	pH	pH	pH	pH	pH	pH	pH	pH	pH
0.0	6.514	6.514	6.605	6.605	6.607	6.607	6.619	6.619	6.621	6.621
0.1	6.512	6.516	6.579	6.634	6.596	6.629	6.600	6.620	6.549	6.629
0.2	6.499	6.543	6.529	6.637	6.561	6.644	6.526	6.636	6.472	6.650
0.4	6.444	6.610	6.475	6.678	6.494	6.654	6.395	6.695	6.418	6.657
0.8	6.357	6.651	6.371	6.712	6.376	6.712	6.289	6.768	6.285	6.690
1.6	6.147	6.717	6.120	6.765	6.161	6.730	6.093	6.852	6.102	6.850
3.2	5.811	6.854	5.785	6.933	5.750	6.951	5.806	6.993	5.730	6.989
6.4	5.328	7.132	5.285	7.271	5.292	7.295	5.323	7.359	5.297	7.390
9.6	4.923	7.514	4.918	7.743	4.794	7.817	4.850	7.767	4.878	7.940
12.8	4.466	8.021	4.362	8.483	4.386	8.715	4.389	8.747	4.494	8.699

Although the differences are not marked they are uniform in showing that the remade milks all showed less buffer effects at low concentration of acid and alkali than were obtained for the natural milk.

Enzyme activity. The remade milks were tested for peroxidase both by the Storch test, using hydrogen peroxide and paraphenylenediamine, and by the guaiac test using hydrogen peroxide and a tincture of gum guaiac. Except for the drum process milk all the samples gave tests which could not be distinguished from raw or pasteurized milk. Both tests were negative for the drum process milk.

Inasmuch as the peroxidase enzyme of milk is destroyed at a temperature of 78° to 82°C., it is obvious that the powders

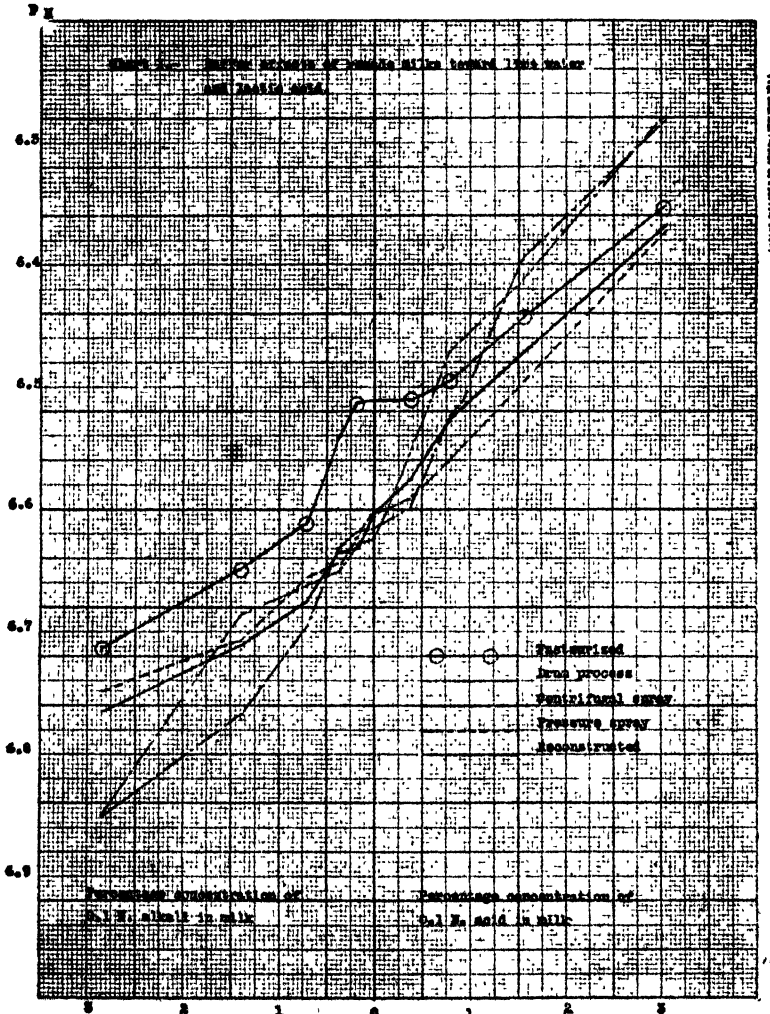


CHART 1

manufactured by the spray process do not attain this temperature while that made by the drum process exceeds this temperature during the manufacture.

SUMMARY

A study of the physical and chemical properties of milk remade from three types of whole milk powder and reconstructed skim milk powder and butter led to the following results:

Remade milk shows the freezing point of fresh milk when made with the proper proportions of powder and water. For the drum process powder this was found to be 10.6 per cent powder, for the centrifugal spray 12.8 per cent powder, for the pressure spray 12.5 per cent powder, and for the reconstructed milk, 9.16 per cent powder and 4.16 per cent butter.

The specific gravity of the remade milks was found to lie within the limits of variation of fresh milk when made up in the proportions which give a normal freezing point.

The viscosity of the remade milks was higher than normal. This was especially true of milk remade from the drum process powder.

The specific electrical conductivity of each type of milk except the drum process was normal when the milks were made up in the proportions which gave a normal freezing point. The drum process milk exhibited a high conductivity which may have been due to the fact that this powder is made from partially skimmed milk.

The milk made from the centrifugal spray powder was the only one which gave a normal cream layer on standing.

The milk made from the centrifugal spray powder showed no deficiency of soluble proteins, but the soluble protein content of the other milks was less than normal. This was especially true of the milk made by the drum method.

The drum process milk and the reconstructed milk failed to coagulate with rennet. The spray process milks showed a rennet coagulability and character of curd somewhat like that of pasteurized milk, the centrifugal spray milk being the nearest to the pasteurized milk in this regard.

At a low concentration of added acid and alkali all the remade milks showed less buffer than normal milk in terms of pH.

Peroxidase was absent from the drum process milk, but was strongly active for the other remade milks examined.

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THE DETERMINATION OF YEASTS AND OIDIA IN CREAM AND BUTTER

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Among the various factors that determine the keeping quality of butter, yeasts and *Oidium lactis* have been considered of importance by many authorities. Jensen (1902) found that pure cultures of *Oidium lactis* produced acid by splitting butter-fat in butter, and that this acid caused rancidity. Rogers (1904) reports the fat-splitting enzymes of certain *torulae* as being responsible in part for the development of acidity in stored butter and suggests that it is reasonable to suppose that they help to produce the so-called "fishy" flavor in butter packed in large vessels. Sayer, Rahn, and Farrand (1909) claim that *Oidium lactis*, because of its fat-splitting qualities, might be one of the causes of rancidity, but no conclusive proof is offered. Thom and Shaw discuss molds as present and offer suggestions for their control. Combs and Eckles (1917), working with both sweet and sour cream, show that *Oidium lactis*, as well as *Penicillium chrysogenum*, when grown in cream previous to pasteurization, exerted a decidedly detrimental effect on the keeping qualities of the butter. This was due to the enzymes carried over into the butter and not to the growth of the mold itself. Eckles (1919) recommends pasteurization at 175° F. to destroy these enzymes and thus insure better keeping quality. Lund (1919) states that butter containing large numbers of yeasts may score high when fresh, but after four months in storage the score will be lower than that of butter containing fewer yeasts. Nicholls (1919) claims that *Oidium lactis* when present is an important factor in the deterioration of butter, and that its enzymes are destroyed only at temperatures above 180°F.

In undertaking to study the presence and function of any group of organisms in butter-making it must be recognized that

many factors may enter into the changes occurring in a lot of cream before churning as well as into the finished butter. Further, it is practically impossible to conduct experiments under conditions which will limit the agencies present to single factors. The results of experiment are, therefore, aggregate effects of several factors rather than evolutions of single factors. Many bacteria survive and multiply in close association with yeasts and *Oidium lactis*. Studies of the presence and effects of particular species must seek to correlate the presence or absence of the particular organism with the characteristic qualities of the butter in series of experiments rather than in isolated tests.

During the summer of 1918, a series of samples of cream and butter were obtained and examined in Denver. The cream used represented the daily receipts of a large creamery, drawing much of its supply from long distances. No attempt to obtain the complete history of each sample was made. The samples were subjected to extensive chemical and bacteriological study for the general purpose of correlating the condition and quality of the cream used with the characteristics determinable in the finished butter. After many experiments the following adaptation of the Breed¹ method for the microscopic count of microorganisms in dairy products was developed.

Cream. For making microscopic slides, the cream is heated to 30°C. and thoroughly mixed. A Breed pipette, calibrated to deliver 0.01 cc. of cream of the average consistency, is used to transfer the sample to the slide, upon which it is spread over 1, 2, or 4 square centimeters according to the quality of the cream under investigation. As much distilled water as necessary is added when spreading over 2 or 4 square centimeters. The method of spreading over a large area as equivalent to dilution is adopted as requiring less time and being more accurate.

Butter. By means of a small butter trier, 3 or 4 cores from different portions of the butter sample are transferred to a 6 by 1 inch test tube, usually about half filling the tube. The butter is melted at 45°C. and allowed to stand at temperatures

¹ Standard Methods for the Bacteriological Examination of Milk, 3d edition, Boston, 1921, pp. 14-16. Published by the American Public Health Association.

between 45° and 40°C. until the fat is separated in a clean layer with mixture of curd and whey or brine at the bottom of the tube. With a sterile 1 cc. pipette the curd and whey are thoroughly mixed by drawing the liquid back and forth in the pipette. Then 1 cc. of this mixture is transferred to a clean watch glass, care being taken to wipe thoroughly all fat from the outside of the pipette with a clean towel before discharging the pipette into the watch glass.² With a Breed pipette 0.01 cc. of the mixture is transferred to a microscopic slide, carefully spread over an area of 1 sq. cm., 2 sq. cm., or 4 sq. cm., depending on the quality of the butter, diluting if necessary with distilled water in order to obtain uniform smears. After drying in the air, extracting with xylol, and fixing with 95 per cent alcohol, the smears are stained with saturated aqueous methylene blue, after which they are washed with water and then with 95 per cent alcohol and dried in the air.

Counting is done with a combination of lens and draw tube length to give a factor of 500,000.³ (See Standard Methods for the Bacteriological Examination of Milk, 3d edition, 1921, pp. 14-16). In enumerating yeasts and oidia each cell, whether separate or as an element in a chain or budding colony, was counted. The number of fields to be counted will depend upon the number of organisms present. It is recommended that 100 fields be counted except in very high count products.

CULTURAL EXAMINATION

Cream was warmed to 30°C., thoroughly mixed by agitation before portions were withdrawn for examination. In the case of butter, four cores were withdrawn from the sample with a sterile trier and placed in a sterile bottle, warmed sufficiently to make the butter soft and mixed thoroughly with a sterile spatula; 10.1 grams of butter were weighed into a sterile porcelain mortar and enough sterile sand was added to give a dry granular mixture when ground up with a sterile pestle. This was then scraped with a sterile spatula into a sterile 500 cc. wide mouth, glass-

² A Petri dish may be used but is not as satisfactory.

³ The diameter of the field should be 0.16 mm. to give a factor of 500,000.

stoppered bottle, 90 cc. of sterile water was added and the bottle was shaken vigorously until a uniform mixture was obtained. From this 1:10 dilution the higher dilutions were prepared.

In examining the colonies on plates, if there was any doubt as to whether these colonies were of bacteria, yeasts or molds, a water suspension was in all cases examined under the microscope. It was found helpful to have a little methylene blue present in the water used for making these suspensions.

In the cultural examination of the samples, the following data were recorded: Counts of yeasts and oidia by the microscopic method, counts of yeasts and oidia in wort agar and in whey agar after five days at 30°C., a microscopic count of total bacteria by the Breed method, a total count of bacteria upon whey agar after five days at 30°C., a total count on wort agar after five days at 30°C., and determinations of the percentages in the following groups: (1) acid organisms; (2) acid coagulating organisms; (3) alkaline organisms; (4) organisms producing no change; (5) peptonizing organisms and (6) organisms which both coagulate and peptonize. The results of microscopic and cultural examination were brought together, and tabulated.

Careful consideration of the tabulated totals obtained from bacteriological examination of freshly made butter indicated that the actual numbers of bacteria present had little significance. The addition of starter introduced so large a factor of the purely acid type of organism as to dominate the bacteriological findings at this stage. The other groups represented only occasionally showed a significant number of organisms. A large number in the peptonizing group occurred only in butter showing very low scores.

The numbers of yeasts and oidia found, however, suggested a possibility of attaching some significance to this determination. The counts of these organisms in the entire series of butter samples were tabulated (table 1). The samples were arranged in the order of butter score, beginning with the highest scoring sample. The corresponding microscopic count of the cream used in making these samples of butter is introduced wherever it was obtain-

TABLE 1

Yeast and oidium counts in butter samples arranged according to commercial score

DESIGNATION	NUMBER	SCORE	MICROSCOPICAL COUNT		CULTURAL COUNT		MICROSCOPIC COUNT IN THE CREAM USED	
			Yeast	Oidium	Yeast	Oidium	Yeast	Oidia
Exp. past.	93	92.7	215,000	<15,000	30	60	435,000	35,000
Exp., starter..	92	92.5	335,000	165,000	7,000	10,000	2,785,000	<15,000
Exp. past.	90	92.0	135,000	<15,000	5	1	1,815,000	<15,000
Exp., cream, untreated...	88	91.5	35,000	<15,000	240	500	800,000	<15,000
Exp., cream, untreated...	91	91.5	435,000	100,000	1,100	7,000	1,685,000	<15,000
Cent., 1st.	19	91.5	815,000	100,000	<10	<10		
Exp., starter..	89	91.0	300,000	<15,000	400	500	2,650,000	<15,000
Exp., neut.	100	90.5	400,000	65,000	50	40	585,000	165,000
Stored 1 yr.	31	90.5	300,000	<15,000	3	0		
Stored 1 yr.	30	90.0	465,000	<15,000	1	1		
Cent., 1st.	13	90.0	985,000	15,000	1,100	<10		
Exp., neut.	99	89.5	50,000	30,000	40	20	1,065,000	265,000
Exp., neut.	39	89.5	55,000	<15,000	1	0	510,000	40,000
Renovated.	15	89.5	650,000	35,000	90,000	1,300		
Exp., neut.	63	89.5	335,000	35,000	4	6	285,000	15,000
Exp., neut.	98	89.0	580,000	230,000	20,000	12,000	3,035,000	450,000
Renovated.	74	88.5	250,000	<15,000	2,300	240		
Exp., neut.	38	89.0	95,000	<15,000	0	0	550,000	20,000
Exp., neut.	35	88.5	110,000	40,000	40,000	2,300	1,720,000	150,000
Cent., 1st.	14	88.5	485,000	100,000	160	10		
Cent., 2d.	20	88.5	1,315,000	35,000	<10	<10		
Exp., starter..	97	88.5	265,000	15,000	20,000	23,000	2,315,000	85,000
Renovated.	86	88.5	35,000	15,000	4,000	400		
Exp., neut.	81	88.5	165,000	<15,000	10	8	750,000	35,000
Exp., neut.	62	88.5	415,000	<15,000	7	3	900,000	<15,000
Exp., neut.	61	88.5	1,150,000	165,000	4,000	4,000	4,265,000	185,000
Exp., neut.	80	88.0	215,000	<15,000	280	40	865,000	<15,000
Exp., neut.	79	88.0	235,000	<15,000	24,000	9,000	3,515,000	385,000
Renovated.	75	88.0	85,000	<15,000	3,500	260		
Renovated.	73	88.0	65,000	<15,000	700	240		
Exp., starter..	36	88.0	425,000	40,000	2,900	400	1,815,000	95,000
Exp., starter..	60	88.0	665,000	200,000	13,000	7,000	2,535,000	315,000
Exp., past.	96	87.5	230,000	50,000	10	10	2,350,000	535,000
Exp. untr. cream.	95	87.5	1,000,000	300,000	14,000	13,000	1,065,000	265,000

Abbreviations: Exp. = experiment; past. = pasteurisation; Cent. = centraliser; neut. = neutralisation; untr. = untreated; Starter-made from cream to which starter was added.

TABLE 1—Continued

DESIGNATION	NUMBER	SCORE	MICROSCOPICAL COUNT		CULTURAL COUNT		MICROSCOPIC COUNT IN THE CREAM USED	
			Yeast	Oidium	Yeast	Oidium	Yeast	Oidia
Exp. untr.								
cream.....	46	87.5	815,000	150,000	7,000	10,000		
Exp., starter..	78	87.0	400,000	165,000	8,000	11,000	2,435,000	550,000
Stored 1 yr...	57	87.0	150,000	50,000	8	1		
Stored 1 yr....	29	87.0	135,000	15,000	0	2		
Stored 1 yr...	28	87.0	85,000	15,000	10	30		
Exp., neut....	25	87.0	115,000	15,000	<10	<10	850,000	35,000
Exp., past....	37	87.5	2,570,000	95,000	0	1	1,720,000	150,000
Exp., starter..	48	87.0	700,000	350,000	70,000	16,000	3,205,000	455,000
Exp. untr.								
cream.....	34	87.0	1,140,000	70,000	3,000	3,200	2,750,000	110,000
Stored 1 yr....	56	87.0	135,000	<15,000	11	0		
Exp. untr.								
cream.....	58	87.0	1,235,000	200,000	4,000	8,000		
Exp. neut....	23	86.7	2,235,000	365,000	34,000	30,000	5,900,000	400,000
Exp., neut....	59	86.5	1,450,000	200,000	130	10	1,135,000	365,000
Exp., past....	77	86.5	365,000	50,000	70	22	4,135,000	750,000
Exp., neut....	51	86.5	150,000	35,000	1	24	700,000	220,000
Stored 1 yr...	41	86.0	20,000	<20,000	0	0		
Cent., 1st....	17	86.5	1,400,000	100,000	1,200	800		
Renovated....	94	87.0	15,000	<15,000	900	40		
Stored 1 yr...	43	86.0	645,000	<20,000	0	0		
Exp., neut....	24	86.0	1,265,000	200,000	500	360	2,285,000	135,000
Stored 1 yr....	53	86.0	35,000	50,000	12	70		
Stored 1 yr...	42	86.0	620,000	70,000	0	0		
Stored 1 yr...	40	85.5	40,000	20,000	0	0		
Exp., neut....	50	86.0	385,000	135,000	3	4	1,140,000	205,000
Exp., past....	47	86.0	215,000	<15,000	50	20	2,475,000	425,000
Stored 1 yr...	52	85.5	65,000	15,000	16	80		
Stored 1 yr...	33	87.5	<15,000	<15,000	?	?		
Stored 1 yr...	32	85.5	15,000	<15,000	2	3		
Exp. untr.								
cream.....	22	85.0	750,000	115,000	11,000	1,400	3,285,000	450,000
Exp., neut....	49	85.5	350,000	50,000	12,000	18,000	1,830,000	260,000
Cent., 2d....	18	85.0	850,000	50,000	160	80		
Stored 1 yr...	45	85.0	550,000	55,000	320	460		
Exp., 3d.....	27	84.5	1,235,000	335,000	<10	<10	2,415,000	400,000
Stored 1 yr...	55	84.5	685,000	385,000	6	0		
Stored 1 yr...	44	84.0	895,000	125,000	160	110		
Exp. untr.								
cream.....	76	84.5	1,900,000	235,000	25,000	14,000	3,300,000	715,000
Stored 1 yr...	54	84.0	1,715,000	285,000	12	0		

TABLE 1—*Concluded*

DESIGNATION	NUMBER	SCORE	MICROSCOPICAL COUNT		CULTURAL COUNT		MICROSCOPIC COUNT IN THE CREAM USED	
			Yeast	Oidium	Yeast	Oidium	Yeast	Oidia
Exp., starter..	26	83.5	800,000	150,000	3,000	21,000	3,235,000	150,000
Packing stock.	69	80.5	2,785,000	385,000	330,000	100,000		
Packing stock.	85	78.5	485,000	50,000	900,000	200,000		
Packing stock.	84	78.0	6,815,000	735,000	300,000	14,000		
Packing stock.	87	77.0	735,000	115,000	410,000	90,000		
Packing stock.	71	77.0	6,135,000	200,000	180,000	180		
Packing stock.	72	76.0	6,965,000	1,000,000	600,000	50,000		
Packing stock.	70	76.0	3,700,000	400,000	190,000	16,000		
Packing stock.	82	75.0	315,000	65,000	320,000	10,000		
Packing stock.	65	75.0	5,765,000	2,750,000	160,000	<10		
Packing stock.	64	75.0	1,785,000	1,185,000	38,000	37,000		
Packing stock.	83	74.5	565,000	400,000	22,000	270		
Packing stock.	16	74.0	1,215,000	1,250,000	6,800,000	330,000		
Grease butter.	68	74.0	1,885,000	115,000	3,600	200		
Packing stock.	67	74.0	1,065,000	300,000	200,000	24,000		
Packing stock.	66	74.0	2,735,000	715,000	120,000	25,000		
Packing stock.	21	73.5	4,750,000	4,015,000	500,000	700,000		

able. The butter investigated included but 3 samples scoring 92 or above. The other samples show progressively such lower scores as to justify the search for agents of deterioration.

From this table, the contrasts between the microscopic counts and the cultural counts in many cases represent the effectiveness of pasteurization of these samples. In general, low-grade samples show high counts of yeasts and oidia. Discrepancies occur frequently enough to indicate that other agents than these groups are capable of causing deterioration. No generalization is possible because this group of samples included too few high-grade products to establish a basis for comparison.

Yeasts and *Oidium lactis* spores are microscopically determinable in butter by the method proposed. The occurrence of these forms in both cream and butter which show signs of deterioration suggest the possibility of correlating the numbers of these organisms in the finished product with conditions in the cream used in manufacture.

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THE STAGE OF LACTATION AS A FACTOR IN THE VARIATION OF THE PER CENT OF FAT IN COW'S MILK

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It is in general known that there is a relation between the stage of lactation and the percentage of fat in milk. Pingree (1) concluded from a study of the advanced registry records of eighteen Guernsey cows that the percentage of butter fat showed, on the average, a tendency to increase throughout the entire lactation period but the increase was most marked during the first five months and during the last month. Van Slyke (2) found that the percentage of fat and protein dropped in the second month of lactation, as compared with the first, and then began to increase, continuing to increase from month to month during the entire period of lactation. In the tenth and eleventh months of lactation, the increase of fat and of proteins is more marked than during the preceding months. Eckles and Shaw (3) found in a study of eleven cows of four breeds that there is a decline in fat during the first three months, followed by a period of from four to five months with little change. From this period on to the end of the lactation period the fat increases rapidly reaching the maximum at the close. They also conclude that there is, in general, some relation between the amount of milk and the per cent of fat. When there is a sudden decline in the amount of milk secreted for any reason, as, for example, during sickness the per cent of fat with few exceptions increases sharply. The same holds true in the rapid decline that occurs in the last portion of the lactation period. Thorndike (4) found that the stage of lactation period makes a cow tend to vary from the general average composition of her milk as follows: First month 89.6, second month 90.3, third 92.4, fourth 96.5, fifth 97.9, sixth 101.2, seventh 103.2, eighth 103, ninth 104.3, tenth 104.9, eleventh 105.3, and twelfth 109.4. Grady (5) presents data on 10 Jersey

and 10 Holsteins showing the effect of the stage of lactation on both the yield of milk, fat, and the per cent of fat. He states that the fat content of the milk varied but little during the first four months. After the fourth month the percentage of fat gradually increased as the lactation advanced. The range in fat content of Jersey milk was somewhat greater than that of the Holstein.

The object of this paper is to present the relation between the stage of lactation and percentage of fat based on a study of 4045 records.

TABLE 1

*Influence of the stage of lactation on the percentage of fat in cows' milk**

MONTH OF LACTATION	GUERNSEY (3763) PER CENT FAT	JERSEY (299) PER CENT FAT	HOLSTEIN (95) PER CENT FAT
1	4.63	4.98	3.24
2	4.59	4.82	3.01
3	4.71	4.88	2.99
4	4.85	5.10	3.02
5	4.97	5.13	3.01
6	5.08	5.26	3.08
7	5.16	5.40	3.11
8	5.22	5.43	3.16
9	5.29	5.50	3.19
10	5.39	5.58	3.27
11	5.49	5.60	3.32
12	5.60	5.73	3.49

* Credit for the calculations involved in deriving the values in table 1 from the records is due the following students: Gerald Petty for the Guernseys, C. A. McCause for the Jerseys, and J. E. Crosby, Jr., for the Holsteins.

The data presented in table 1, in the form of average percentages of fat, were derived from a study of the records of 3763 Guernsey Advanced Register tests, 299 Jersey Register of Merit tests, and 95 Holstein Advanced Register tests.

The Guernsey records were secured from the Advanced Register of Guernsey Cattle. The records published by the Guernsey Cattle Club are exceptionally valuable for a study of this nature as the itemized monthly productions of milk and fat as well as the average per cent of fat are published with each completed record. The animals used were representative with

respect to their ability as producers. The animals were scattered over the United States to some extent but the East and Central West where most of the Guernsey tests are being made was especially well represented.

Influence of the Advance of Lactation on the per cent
of Fat in Cows' Milk.

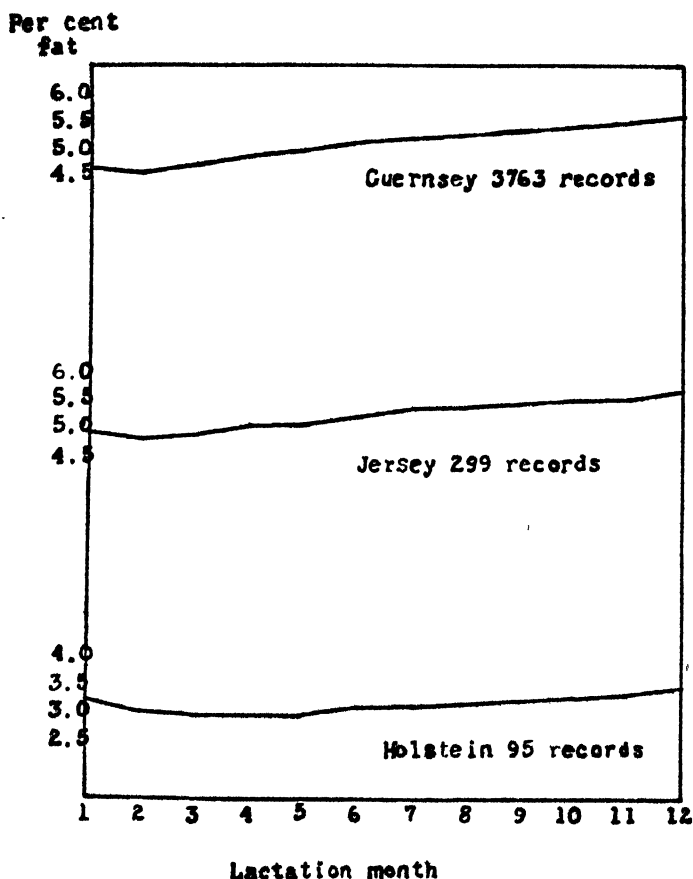


FIG. 1

The Jersey records were secured from the American Jersey Cattle Club. They consist of the records of Jersey cattle tested in Missouri during the past few years. All completed records available were used in this study so that animals of varying

producing ability and handled under varying conditions from the standpoint of management, feeding, etc., are represented.

The Holstein records were secured from the records of the Holstein herd owned by the University of Missouri. Here again the records used were of animals of varying productive ability and age but were made under similar herd conditions.

The data presented in table 1 and figure 1 shows that the per cent of fat during the first month is high and that there is a gradual decline in the per cent of fat, reaching its lowest point the second or third month. Following the low period there is a gradual increase in the per cent of fat. During the last month there is a more rapid increase in the per cent of fat up to the time the flow of milk ceases. It is undoubtedly true that there is some relation between the amount of milk produced and the per cent of fat (3-5). The sharp and more extended decline of the Holsteins as compared to the other breeds is probably due to the fact that the Holstein cows are generally tested for seven day records during the first month. This is usually as soon after calving as is permitted by the rules in order to secure the somewhat higher per cent of fat that may be expected from a cow in high condition at calving time. While it is customary to have cows that are put on yearly test in good condition, the same attention is not given to having them excessively fat. Eckles (6) has shown that the per cent of fat in milk can be influenced to a marked extent for the first twenty to thirty days by the fatness of the animal at parturition. This influence appears to extend in some cases in a less degree for at least three months.

SUMMARY

It is shown that under conditions such as ordinarily prevail in the management of dairy cattle, and independent of the season of the year or character of the diet, the stage of lactation does have an influence on the per cent of fat in cows' milk. There is a noticeable decline in the per cent of fat from the first month to the second and in some cases this influence continues in a less degree for at least three months. Following the low period there

is a gradual increase in the per cent of fat which becomes more pronounced during the last month of lactation.

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COCONUT MEAL, GLUTEN FEED, PEANUT MEAL AND SOYBEAN MEAL AS PROTEIN SUPPLEMENTS FOR DAIRY COWS

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The dairy farmer of the corn belt can grow all the roughages necessary for his cattle and the best results are obtained when corn silage and clover or alfalfa hay are produced for this purpose. Grains of low protein content, such as corn and oats, are also easily produced in this section and the feeds that have to be purchased most generally are the concentrates of high protein content.

Much work has already been done in comparing feeds of this class so far as their value for milk production is concerned but in view of the newer knowledge of nutrition which demonstrates that the value of a feed may depend to a large degree on the nature of the other feeds with which it is fed it was felt that some additional work of this character need not necessarily be a duplicate of previous work. The basal rations used were constituted of materials commonly grown on corn belt farms, while the concentrates studied were ones on which little experimental data is available. The feeds studied were soybean meal, peanut meal, corn gluten feed and coconut meal, and they were compared with old process linseed oil meal.

Coconut meal. This feed, frequently known as copra meal, has been in use for a number of years on the Pacific Coast and during the war it seemed that the interruption of Germany's trade caused larger quantities of this feed to be shipped to the United States. There is now a fair trade in this commodity and it is handled by several distributing houses in the middle west. It is hardly probable however that large quantities will ever be available in this section.

Gluten feed. The increase in the manufacture of starch and glucose from corn has increased considerably in recent years,

and so larger amounts of the by-product gluten feed are becoming available. Much of this feed is produced in the corn belt, and though the greater part of it was for a long time shipped East, a considerable quantity of it is now used in this section.

Peanut meal. The growth of the peanut industry has been rapid and large amounts of the nuts are used for the manufacture of oil. Where the meal, that is left after the extraction of the oil, comes from hulled nuts, a feed of exceptionally high protein content is obtained. The meal from the unhulled nuts is known as peanut feed and is of considerably lower value.

Soybean meal. For a number of years soybean meal has been imported in considerable quantities to the Pacific Coast states from Japan, Manchuria and other regions in the far East. Freight rates prohibited the use of this feed in the middle west. Within recent years however the growing of soybeans in the South has increased rapidly and, as a large quantity of the beans is used for oil extraction, the soybean meal which is obtained as a by-product has been coming on the market in considerable quantities.

RESUMÉ OF PREVIOUS WORK

The compilation of a summary of work on a problem of this character tends to lead to confusion, as few direct comparisons of the feeds under consideration have been made and the conditions under which the trials have been made vary greatly. Consequently, it is necessary to consider the concentrates separately and also to give some attention to the work with cottonseed meal and oil meal in direct comparison as these two feeds have generally been the basal materials with which the newer concentrates have been compared.

Cottonseed meal vs. linseed meal

It was found by Waters and Hess (12) that cottonseed meal gives more milk and about the same amount of butterfat as oil meal, while Hills (5), Hunziker and Caldwell (7) and Michels (9) obtained better results with cottonseed meal than with linseed meal.

Cottonseed meal vs. other protein supplements

Both Scott (11) and Fitzpatrick (3) found coconut meal to be of about the same value as cottonseed meal, while Hunziker and Caldwell (7) stated that cottonseed meal was more economical than gluten feed. From their work Ewing and Ridgway (2) concluded that peanut meal was less valuable than cottonseed meal, while Gilchrist (4) found that soybean cake was slightly superior to cottonseed cake for milk production.

Linseed meal vs. other protein supplements

It was found by Hunziker and Caldwell (7) that gluten feed was more economical than linseed meal, while Hansen (6) found soybean cake and linseed cake to be of about equal value.

Gluten feed vs. coconut meal

It is stated by Lindsey (8) that coconut meal is of about the same value as gluten feed.

Gluten feed and peanut meal vs. no protein supplement

On replacing wheat bran and corn meal with an equal weight of gluten feed, Cooke (1) obtained an increase of 15 per cent in milk and 16 per cent in fat yield, while Pott (10) states that good results are obtained with peanut meal.

In briefly summarizing these results of the limited amount of work reported it may be said that on the whole cottonseed meal appears to give the best returns with linseed oil meal as a close second. There would also appear to be little difference in the values of the other feeds when compared with oil meal or with each other but that the addition of a supplement high in protein to a ration lacking such a supplement will increase milk and butterfat production.

EXPERIMENTAL WORK

The work reported here consisted of two trials of 150 days each in the first of which peanut meal and soybean meal were compared with old process linseed oil meal, while in the second trial coconut meal and gluten feed were compared with the linseed meal.

The analysis of the protein supplements used are given and for purposes of comparison they are arranged in order of their content of crude protein. All feeds were in good condition when used so any differences found cannot be attributed to variations in quality.

It will be noted that the peanut and soybean meals contain about 8 to 9 per cent more crude protein and the gluten feed and coconut meal 11 to 15 per cent less crude protein than does the oil meal.

TABLE I
Composition of protein supplements

FEED	PEANUT MEAL	SOYBEAN MEAL	LINSEED OIL MEAL O. P.	GLUTEN FEED	COCONUT MEAL
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
Moisture.....	6.78	8.79	8.02	8.64	9.55
Dry matter.....	93.22	91.21	91.98	91.36	90.45
Crude protein.....	44.44	43.65	34.90	23.03	19.31
Nitrogen free extract.....	24.73	29.00	33.64	53.78	44.22
Crude fiber.....	10.63	6.12	10.82	8.07	11.71
Ether extract.....	8.76	6.28	7.32	4.21	8.73
Ash.....	4.66	6.16	5.30	2.27	6.48

Comparison of peanut meal and soybean meal with linseed meal

In this trial three cows were used. Previous to this work they had been handled with the general herd. During the experiment they were milked twice daily, allowed to exercise in yards whenever the weather permitted. They had salt before them at all times. They were watered morning and evening and were weighed before and after watering, so that a record of water consumption might be obtained. Hay, grain, and silage were each fed twice daily, the grain being fed on top of the silage. Some information concerning the animals used is given in table 2 and where necessary it is calculated to November 18, 1918, the day on which the experiment started.

The basal ration fed consisted of corn silage, alfalfa hay and a grain mixture of 2 parts cracked corn, 2 parts ground oats and 1 part wheat bran by weight. The roughages were fed according

TABLE 2
Animals used

	COW 253, GRADE GUERNSEY	COW 267, GUERNSEY	COW 284, JERSEY
Age, years.....	4½	4	3½
Fresh, days.....	72	140	109
Bred, days.....	0	0	0
Previous lactations.....	3	2	2

TABLE 3
Live weight, feed consumption, milk and butterfat production

PERIOD NUMBER	COW NUMBER	AVER- AGE LIVE WEIGHT	TOTAL FEED CONSUMPTION				TOTAL PRODUCTION	
			Corn silage	Alfalfa hay	Grain mixture	Protein supple- ment	Milk	Fat
		pounds	pounds	pounds	pounds	pounds	pounds	pounds
I. Oil meal.....	253	1019	600	160	120	60	353.6	16.99
	267	871	500	160	120	60	450.6	23.65
	284	888	500	160	140	70	481.2	28.24
	Group	926	1600	480	380	190	1285.0	68.88
II. Peanut meal.....	253	1035	600	160	120	60	332.7	16.22
	267	887	500	160	120	60	434.4	22.15
	284	890	500	160	140	70	465.1	28.60
	Group	937	1600	480	380	190	1232.2	66.97
III. Oil meal.....	253	1072	600	160	120	60	339.4	15.86
	267	906	499	160	120	60	426.1	21.20
	284	896	500	160	140	70	428.6	25.51
	Group	953	1599	480	380	190	1194.1	62.57
IV. Soybean meal.....	253	1070	500	160	100	50	326.3	14.93
	267	927	496	160	120	60	414.2	20.61
	284	891	500	160	120	60	405.1	24.81
	Group	963	1496	480	340	170	1145.6	60.35
V. Oil meal.....	253	1059	400	160	80	40	308.4	13.80
	267	937	500	160	120	60	417.4	20.04
	284	897	500	160	100	50	393.0	22.99
	Group	964	1400	480	300	150	1118.8	56.83

to the weight and capacity of the animals while the grain ration was controlled largely by production. Two parts of the grain mixture mentioned were fed along with one part, by weight, of the protein supplement being considered.

The experiment, of 150 days duration, was divided into 5 periods of 30 days each. The basal ration was fed throughout all periods while the protein supplements were old process linseed

TABLE 4
Peanut meal and soybean meal vs. old process linseed oil meal

PERIOD	AVERAGE LIVE WEIGHT	TOTAL PRODUCTION			TOTAL FEED CONSUMPTION			
		Milk	Fat	Fat	Corn silage	Alfalfa hay	Grain mixture	Protein supple- ment
Peanut meal vs. oil meal								
	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>per cent</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>
Oil meal.....	940	1239.6	65.72	5.30	1600	480	380	190
Peanut meal.....	937	1232.2	66.97	5.43	1600	480	380	190
Increase.....			1.25	0.13				
Decrease.....	3	7.4						
Increase percent..			1.9	2.5				
Decrease percent.	0.3	0.6						
Soybean meal vs. oil meal								
	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>per cent</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>
Oil meal.....	959	1156.5	59.70	5.16	1500	480	340	170
Soybean meal.....	963	1145.6	60.35	5.27	1496	480	340	170
Increase.....	4		0.65	0.11				
Decrease.....		10.9			4			
Increase percent..	0.4		1.1	2.1				
Decrease percent.		0.9			0.3			

oil meal in the first, third, and fifth periods, peanut meal in the second period and soybean meal in the fourth.

The first 10 days of each 30-day period was looked on as a preliminary period and the comparisons are consequently obtained from the remaining 20 days of each period.

In order that the protein supplements being studied may be compared with the oil meal, the results obtained with the supplement under consideration are compared with the average for the oil meal periods preceding and following it.

Throughout the periods where peanut meal and soybean meal were compared with the old process linseed oil meal the animals remained very uniform in live weight and the only variation in feed consumption was a difference of four pounds of silage between the soybean meal period and oil meal check periods—a difference which is negligible so far as its effects on the ultimate results are concerned.

The variations in production are also very small—indicating no practical difference in the value of the soybean meal and peanut meal for production purposes, when compared with oil meal.

Comparison of gluten feed and coconut meal with linseed meal

The outline for this work was exactly the same as that for the investigation with the peanut and soybean meal, with the ex-

TABLE 5
Animals used

	COW 280, JERSEY	COW 298, GRADE GUERNSEY	COW 301, GRADE GUERNSEY	COW 323, GRADE HOLSTEIN
Age, years.....	3½	3	3	2½
Fresh, days.....	200	166	239	180
Bred, days.....	12	40	15	57
Previous lactations.....	1	1	0	0

ception that four cows were used. The data concerning these cows is outlined in table 5, and where necessary, calculated November 18, 1918, the day on which the work started.

The basal ration used and the methods of feeding employed have already been outlined in the other part of the work and the results are summarized in similar form.

In this part of the work also great uniformity in the live weights of the animals was maintained. Slight variations in feed consumption did occur but all are negligible with the possible exception of decreases of 3.3 per cent in the basal grain ration and 6.5 per cent in the protein supplement consumed during the coconut meal period. This decrease was largely due to the relatively unpalatable nature of the coconut meal when compared

with oil meal which was exceptionally palatable to all of the animals used in the experiment.

TABLE 6
Live weights, feed consumption, milk and butterfat production

PERIOD NUMBER	COW NUMBER	AVERAGE LIVE WEIGHT	TOTAL FEED CONSUMPTION				TOTAL PRODUCTION	
			Corn silage	Alfalfa hay	Grain mixture	Protein supplement	Milk	Fat
		<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>
I. Oil meal.....	289	773	400	120	120	60	342.8	21.50
	298	859	400	120	120	60	283.5	15.38
	301	779	400	120	120	60	369.1	19.83
	323	984	400	120	120	60	378.6	15.90
	Group	849	1600	480	480	240	1374.0	72.61
II. Gluten feed.....	289	778	400	120	120	60	323.9	21.87
	298	901	398	120	120	60	276.1	15.25
	301	799	400	120	120	60	348.9	19.88
	323	1000	400	120	120	60	345.6	15.03
	Group	870	1598	480	480	240	1294.5	72.03
III. Oil meal.....	289	792	400	120	120	60	291.9	19.99
	298	928	400	120	120	60	249.4	13.84
	301	814	400	120	120	60	342.5	18.50
	323	1018	370	114	111	51	290.6	13.87
	Group	888	1570	474	471	231	1174.4	66.20
IV. Coconut meal.....	289	811	398	113	103	43	255.8	18.75
	298	962	397	119	99	49	235.7	14.14
	301	836	400	120	120	60	317.5	18.41
	323	1020	393	117	80	40	270.1	13.46
	Group	907	1588	469	402	192	1079.1	64.76
V. Oil meal.....	289	819	400	120	120	60	281.8	19.23
	298	964	398	119	80	40	212.2	12.15
	301	827	400	120	120	60	347.1	17.88
	323	1014	400	120	40	20	222.3	9.46
	Group	906	1598	479	360	180	1063.4	58.72

When the production comes to be considered it is found that again there is little apparent difference in the value of the concentrates considered as protein supplements as the greatest difference in milk or butterfat production did not reach 4 per

TABLE 7

Gluten feed and coconut meal vs. old process linseed oil meal

PERIOD	AVERAGE LIVE WEIGHT	TOTAL PRODUCTION			TOTAL FEED CONSUMPTION			
		Milk	Fat	Fat	Corn silage	Alfalfa hay	Grain mixture	Protein supple- ment
Gluten feed vs. oil meal								
	pounds	pounds	pounds	per cent	pounds	pounds	pounds	pounds
Oil meal.....	869	1274.2	69.42	5.45	1585.0	477.0	476.0	236.0
Gluten feed.....	870	1294.5	72.03	5.56	1598.0	480.0	480.0	240.0
Increase.....	1.0	20.3	2.61	0.11	13.0	3.0	4.0	4.0
Decrease.....								
Increase percent..	0.1	1.6	3.8	2.0	0.8	0.6	0.8	1.6
Decrease percent								
Coconut meal vs. oil meal								
	pounds	pounds	pounds	per cent	pounds	pounds	pounds	pounds
Oil meal.....	897	1118.9	62.46	5.58	1584	477	416	205
Coconut meal.....	907	1079.1	64.76	6.00	1588	469	402	192
Increase.....	10		2.30	.42	4			
Decrease.....		39.8				8	14	13
Increase percent..	1.1		3.7	7.5	0.3			
Decrease percent		3.6				1.7	3.3	6.3

TABLE 8

Market value of feeds used

FEED	VALUE PER TON
Corn silage.....	\$ 7.00
Alfalfa hay.....	25.00
Cracked corn.....	25.00
Ground oats.....	30.00
Wheat bran.....	40.00
Linseed oil meal O. P.....	65.00

cent. It should be noted however that relatively the least milk and the highest yield of butterfat as well as the highest butterfat content in the milk was obtained when coconut meal was fed and it was not consumed in as large quantities as was the linseed meal.

Economy of production

Of greater importance than small changes in production is economy of production as it is on this point that the dairy business hinges. In arriving at some idea of the relative value of the protein concentrates considered it will therefore be necessary to give some consideration to cost of production. In deter-

TABLE 9
Value and feed cost of product and value of protein supplement

	TOTAL YIELD		VALUE OF PRODUCT	FEED COST LESS PROTEIN SUPPLEMENT				VALUE OF 1 TON OF SUPPLE- MENT
	Milk	Fat		Corn silage	Alfalfa hay	Grain mixture	Total	
Peanut meal vs. oil meal								
	<i>pounds</i>	<i>pounds</i>						
Oil meal.....	1239.6	65.72	\$57.74	\$5.60	\$6.00	\$5.70	\$17.30	\$65.00
Peanut meal.....	1232.2	66.97	58.53	5.60	6.00	5.70	17.30	66.00
Soybean meal vs. oil meal								
Oil meal.....	1156.5	59.70	52.76	5.25	6.00	5.10	16.35	65.00
Soybean meal.....	1145.6	60.35	53.10	5.24	6.00	5.10	16.34	66.00
Gluten feed vs. oil meal								
Oil meal.....	1274.2	69.42	60.64	5.55	5.96	7.14	18.65	65.00
Gluten feed.....	1294.5	72.03	62.64	5.59	6.00	7.20	18.79	67.00
Coconut meal vs. oil meal								
Oil meal.....	1118.9	62.46	54.29	5.54	5.96	6.23	17.73	65.00
Coconut meal.....	1079.1	64.76	55.49	5.56	5.86	6.02	17.44	72.00

mining this the costs of all feeds except the protein supplements studied are taken at market value and the difference between the value of the products and the cost of all feeds except the protein supplement is taken as a measure of the relative value of the supplement for milk production.

The value of 70 per cent per pound is taken for the butterfat and \$1.00 per 100 pounds for the skim milk and so with a value of \$65 per ton for the old process linseed oil meal it is not difficult to get the relative values of the other protein supplements.

When the results are compared it is noticeable that all of the

protein supplements with the exception of coconut meal have a very similar value. Taking old process linseed oil meal at \$65 per ton and using the method outlined, values of \$66, \$66, \$67 and \$72 per ton are obtained for peanut meal, soybean meal, gluten feed and coconut meal respectively. Apparently therefore soybean meal, peanut meal and gluten feed are of about the same value as linseed meal and coconut meal has a higher value than any of the others.

The results obtained are quite comparable with those found in previous work. There seems to be little doubt about the relative values of all of the feeds except coconut meal. It is not very palatable but seems to be quite valuable for milk production though the results obtained in its use have been at times contradictory.

Palatability of protein supplements

The palatability of all of the feeds under discussion has already been mentioned in another connection and during this work coconut meal was apparently the least palatable. The old process linseed oil meal was the most palatable, followed closely by soybean meal, while gluten feed and peanut meal ranked considerably lower.

None of the feeds appeared to have any specific effects on the animals as all kept in good condition throughout the work.

SUMMARY

The following deductions from the trial of protein supplements reported here are probably justified:

1. Old process linseed oil meal, peanut meal, soybean meal and gluten feed are probably of about equal value as protein supplements in the ration of the dairy cow.
2. Coconut meal may have a higher value, as a protein supplement, than the feeds mentioned, even though it has a lower content of total protein. The few pieces of work done on this problem are contradictory in character however.
3. In palatability the feeds studied rank as follows: linseed oil meal, soybean meal, gluten feed, peanut meal and coconut meal.

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FAT ANALYSIS OF MILK POWDER

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Up to the present time the Association of Official Agricultural Chemists has not designated an official method for determining fat in milk powders. The Roesse-Gottlieb method, however, is generally accepted as the best method available for this purpose. Probably because of the fact that this Association has not officially adopted a method for determining fat in milk powder, and because of the urgent need for a method which is entirely reliable, numerous modifications of existing methods have been proposed. Some of these modifications have involved an extraction from an acid solution instead of an alkaline solution as is used in the regular Roesse-Gottlieb procedure; others have involved modifications of the Babcock test, whereby the method could be adopted for factory use.

Biesterfeldt and Evenson (1) advocate the use of weak acetic acid, claiming this reagent allows extraction of fatty acids which may be present and which are not extracted from an alkaline medium. They claim the Roesse-Gottlieb method gives an average of 0.04 per cent too low for condensed milk, and further state that for milk powders, the results may be even lower.

Harding and Parker (2) claim that more fat is extracted from evaporated milk by the use of 25 per cent acetic acid, carbon tetrachloride and petroleum ether than by the reagents used in the regular Roesse-Gottlieb method. Biesterfeldt and Evenson (1), however, think that the higher results obtained by this method are due to the use of rubber stoppers in contact with the reagents.

Mohs (3) recommends a preliminary treatment of milk powder with hydrochloric acid and subsequent extraction in the Soxhlet apparatus with ethyl ether. He states that all of the fat is not

¹Former Laboratory staff members participating in these comparative analyses were, A. H. Green and P. A. Bunce.

removed from milk powder by simple extraction with ethyl ether, and that there appears to be a decrease in fat from old powders due to absorption of the fat by the coagulated protein.

Phillipps (4) recommends extraction with trichlorethylene for rapid extraction of the fat.

Redmond (5) has recommended a modification of the Babcock test for factory use, the results from which are claimed to be from 0.1 per cent to 0.2 per cent higher than by any ether extraction method and 3 per cent higher than the regular Babcock method.

Roop (6) recommends the use of dilute sulphuric acid followed by digestion in the steam bath in the place of ammonia as used in the Roesse-Gottlieb method for fat in dried buttermilk.

It seems to be recognized that a simple ether extraction in the Soxhlet apparatus is distinctly inferior to the Roesse-Gottlieb method for liquid milk, evaporated, condensed or powdered milks. The majority of proposed modifications of methods have sought to improve the efficiency and reliability of the Roesse-Gottlieb method. These modifications as well as the simple Soxhlet extraction method have but little recognition at the present time for determining fat in milk powder, although the Soxhlet method is quite frequently used for this purpose.

The inadequacies of a simple ether extraction for determination of butter fat in milk products were no doubt widely recognized after the official adoption by the Association of Official Agricultural Chemists at the 32nd Annual Conference, November 15, 1915, of the Roesse-Gottlieb method for fat in milk and condensed milk, both sweetened and unsweetened. At the same time it was recommended by associate referee Hortvet (7), that a further study be made of this method in the analyses of ice cream, milk powders, malted milks and milk chocolates. This recommendation was, no doubt, stimulated by the relative accuracy and uniformity of results from evaporated and condensed milks. The results reported from a single sample of evaporated milk on which forty-five analyses were made by nineteen different analysts showed a maximum variation of 0.29 per cent. From two samples of sweetened condensed milk

analyzed by the same collaborators and involving twenty-five to twenty-seven determinations on each sample, the maximum variations were 0.21 per cent and 0.17 per cent.

At the Association meeting held November 20, 1917, Hortvet (8) reported results of collaborative work on analyses for fat in dried skimmed milk by the official Roese-Gottlieb method and by an acid extraction modification of the same. The results from the official method reported from a single sample on which twenty-five analyses were made by nine different analysts showed a maximum variation of 0.33 per cent on a powder containing 1.13 per cent fat as an average of all determinations. The results reported from the acid extraction modification showed a maximum variation of 0.65 per cent on the same sample. On the basis of these results it was recommended that further study be made of the Roese-Gottlieb method as applied to dried milk products containing a high as well as a low butter fat content.

COMPARISONS OF RESULTS FROM DIFFERENT METHODS

During our investigational and control work connected with desiccated milk, it has been desirable to make comparisons of different methods used for determining the butter fat content of this product. The results reported herein were obtained during a period extending over two years from dried milk made by the Just double roller process. The great majority of results, unless otherwise designated, were made with the Mojonnier apparatus which is considered a practical adaptation of the chemical principles of the Roese-Gottlieb method for routine factory analysis; those determinations made by this procedure will be hereinafter designated as the Roese-Gottlieb (Mojonnier) method.

VARIATIONS IN RESULTS OBTAINED BY THE ROESE-GOTTLIEB (MOJONNIER) METHOD

One of the first studies to be made in connection with this work on reliability of methods was for the purpose of ascertaining normal variations between duplicate determinations made by the Roese-Gottlieb (Mojonnier) method. The duplicate deter-

minations made by different analysts on samples of freshly made milk powder are shown in table 1. Since the two determinations on any one sample were made within short intervals, thereby eliminating the possibility of any discrepancy due to variable moisture content, the results are expressed on the original powder basis and not on the moisture-free basis. From the results shown in this table, the maximum variation between duplicate determinations by the Roese-Gottlieb (Mojonnier) method is 0.26 per cent; the minimum variation is zero; and the average variation is 0.099 per cent.

TABLE 1
Results of duplicate determinations for fat in milk powder, Roese-Gottlieb (Mojonnier) method.

SAMPLE NUMBER	DETERMINATION NUMBER 1, PER CENT FAT	DETERMINATION NUMBER 2, PER CENT FAT	DIFFERENCE, PER CENT FAT
1	23.93	24.19	0.26
2	24.47	24.51	0.04
3	26.55	26.55	0.00
4	31.12	31.12	0.00
5	28.12	28.02	0.10
6	25.68	25.47	0.21
7	27.21	27.39	0.17
8	27.07	26.95	0.12
9	28.33	28.29	0.04
10	30.09	30.05	0.04
11	26.21	26.32	0.11
12	19.75	19.84	0.09
13	53.51	53.65	0.14
14	55.00	54.93	0.07
Average.....			0.099

VARIATIONS IN RESULTS OBTAINED BY THE ROESE-GOTTLIEB (MOJONNIER) METHOD FROM FRESH AND OLD MILK POWDER

In order to determine whether or not the ageing of milk powder introduced a factor which would tend to cause discrepancies in the results from the Roese-Gottlieb (Mojonnier) method, several samples of powder made from part skimmed milk and from whole milk were analyzed while fresh and after various storage periods;

moisture determinations were made on each sample at the time of analysis and the fat percentages converted to the moisture-

TABLE 2

Results from Roesse-Gottlieb (Mojonnier) method on part skimmed milk powder while fresh and at age of two months

SAMPLE NUMBER	FRESH	AFTER TWO MONTHS
	<i>per cent fat</i>	<i>per cent fat</i>
1	12.52	12.23
2	13.07	12.73
3	11.37	11.24
4	11.82	11.91
5	11.28	11.23
6	12.47	12.55
7	12.70	12.49
8	12.24	12.54
9	12.58	12.42
10	12.18	12.25
Average.....	12.22	12.15

TABLE 3

Results from Roesse-Gottlieb (Mojonnier) method on part skimmed milk powder while fresh and at age of three months

SAMPLE NUMBER	FRESH	AFTER THREE MONTHS
	<i>per cent fat</i>	<i>per cent fat</i>
1A	12.70	12.69
2A	12.16	12.39
3A	12.58	12.60
4A	12.39	12.47
5A	12.27	12.18
6A	12.29	12.35
7A	12.39	12.14
8A	12.46	12.65
9A	12.23	12.34
10A	12.63	12.56
Average.....	12.41	12.46

free basis. The results of these determinations are shown in tables 2 to 5 inclusive.

In tables 2 to 5 inclusive there is opportunity to compare results from two determinations on thirty-five different samples, one

determination in each case being made while the powder was still fresh and the other when the powder was from two to six months old. The maximum variation between two determinations on the same sample is 0.41 per cent; the minimum is 0.01 per cent; and the average is 0.17 per cent. The results of these

TABLE 4

Results from Roesse-Gottlieb (Mojonnier) method on part skimmed milk powder while fresh and at age of six months

SAMPLE NUMBER	FRESH	AFTER SIX MONTHS
	<i>per cent fat</i>	<i>per cent fat</i>
1B	12.82	13.08
2B	13.76	13.48
3B	12.33	12.18
4B	12.14	12.07
5B	13.23	12.84
Average.....	12.85	12.73

TABLE 5

Results from Roesse-Gottlieb (Mojonnier) method on whole milk powder while fresh and at age of four months

SAMPLE NUMBER	FRESH	AFTER FOUR MONTHS
	<i>per cent fat</i>	<i>per cent fat</i>
1	26.72	26.56
2	27.34	27.32
3	27.76	27.41
4	27.09	27.16
5	27.44	27.82
6	27.41	27.23
7	26.31	26.51
8	27.93	28.25
9	27.48	27.81
10	27.79	27.38
Average.....	27.32	27.29

determinations show wider variations than was found in the samples recorded in table 1. This is undoubtedly due, to some extent at any rate, to the fact that the results from these four series containing thirty-five samples are all expressed on the moisture-free basis, the wider variations being explained in part,

as the result of analytical discrepancies in the moisture determinations.

In considering the question of the extraction of less fat from old powder than from fresh, there were nineteen samples from which less fat was extracted after ageing; the average difference was 0.183 per cent. The remaining sixteen samples gave higher results after ageing; the average difference was 0.154 per cent. Although the average difference between fresh and old samples shows that 0.029 per cent more fat was extracted from the former, this is not considered as conclusive evidence that this method of analysis is less reliable for old powders.

ROESE-GOTTLIEB (MOJONNIER) AND SIMPLE ETHER EXTRACTION METHODS COMPARED

Although the simple ether extraction method with the Soxhlet apparatus is not generally recognized as an efficient method for the determination of fat in milk powder, it is, however, frequently used in commercial laboratories for the purpose of determining the fat content of different consignments of this and other food products carrying a guaranteed fat content. In order to obtain comparative data between the two methods, parallel determinations were made on powder from part skimmed milk and from whole milk when the powder was fresh and after a storage period of four months. In the ether extraction method the extraction was carried out for seven hours. These results which are shown in tables 6 and 7 are all expressed on the moisture-free basis.

From the four different comparisons between the Roese-Gottlieb (Mojonnier) and ether extraction methods which are possible in tables 6 and 7, there was on an average 0.22 per cent more fat extracted by the former method. Each group of parallel determinations shows a similar relationship between the two methods.

Again considering the variations in results between individual determinations on the same sample as shown by both of these methods, it was found that from the Roese-Gottlieb (Mojonnier) method there was a maximum variation of 0.49 per cent; a minimum of 0.01 per cent; and an average of 0.143 per cent. From the simple ether extraction method there was a maximum

variation of 1.29 per cent, a minimum of 0.05 per cent and an average of 0.369 per cent. In these comparisons as in those

TABLE 6

Rosse-Gottlieb (Mojonnier) and simple ether extraction methods compared on part skimmed powder, fresh and after four months

SAMPLE NUMBER	FRESH		AFTER FOUR MONTHS	
	Rosse-Gottlieb	Soxhlet	Rosse-Gottlieb	Soxhlet
	<i>per cent fat</i>	<i>per cent fat</i>	<i>per cent fat</i>	<i>per cent fat</i>
1	12.46	12.01	12.49	12.21
2	12.24	12.34	12.64	12.18
3	12.45	12.36	12.94	12.62
4	12.68	12.13	12.60	13.42
5	12.27	12.19	12.18	11.73
6	12.29	11.99	12.35	11.88
7	12.39	11.96	12.45	12.25
8	12.46	11.88	12.65	12.96
9	12.23	12.06	12.34	12.11
10	12.63	12.33	12.56	12.27
Average.....	12.41	12.12	12.52	12.36

TABLE 7

Rosse-Gottlieb (Mojonnier) and simple ether extraction methods compared on whole milk powder, fresh and after four months

SAMPLE NUMBER	FRESH		AFTER FOUR MONTHS	
	Rosse-Gottlieb	Soxhlet	Rosse-Gottlieb	Soxhlet
	<i>per cent fat</i>	<i>per cent fat</i>	<i>per cent fat</i>	<i>per cent fat</i>
1	26.98	26.33	26.92	26.42
2	26.81	26.75	26.56	26.45
3	27.30	27.09	27.32	27.45
4	27.41	27.43	27.28	27.97
5	27.06	27.21	27.16	26.75
6	27.51	27.39	27.32	26.91
7	27.14	27.49	27.23	26.96
8	26.77	25.73	26.51	26.11
9	26.92	26.28	26.77	26.20
10	27.37	27.29	27.38	27.14
Average.....	27.12	26.89	27.04	26.88

already recorded there is no conclusive evidence that less fat is extracted from old powder than from fresh, regardless of the method.

EXTRACTION FROM AN ACID MEDIUM

Since it has been claimed that more fat is extracted from an acid solution than from an alkaline solution, comparisons were made between one of these methods and the Roesse-Gottlieb (Mojonnier) method. The method used was the one recommended by Roop (6) for dried buttermilk in which powder is digested in dilute sulphuric acid (1:9) in the steam bath for thirty

TABLE 8

Roesse-Gottlieb (Mojonnier) and alkaline and acid extraction methods compared

SAMPLE NUMBER	ACID EXTRACTION	ALKALINE EXTRACTION
	<i>per cent fat</i>	<i>per cent fat</i>
1	27.21	27.85
2	27.47	28.72
3	27.08	26.40
4	26.10	25.73
5	26.51	26.37
6	26.15	25.56
7	26.15	25.46
8	25.49	25.26
9	25.69	25.75
10	26.29	25.70
11	27.46	25.91
12	27.01	26.26
13	26.18	25.57
14	25.96	25.71
15	25.98	25.33
16	25.75	26.05
17	24.52	25.09
18	25.51	26.14
19	26.12	26.01
Average.....	26.24	25.94

minutes prior to extraction. This method as applied to the Mojonnier manipulation differs only in the fact that in the latter method ammonium hydroxide is used to digest the casein, whereas digestion is accomplished by weak acid and heat in the former method. The results obtained from these two methods are shown in table 8. The figures show that out of 19 comparisons, 14 are higher from the acid extraction method, and that the average difference is 0.30 per cent higher from this latter method.

ROESE-GOTTLIEB (MOJONNIER) AND REDMOND METHODS
COMPARED

The results from the Redmond method, in which a given weight of powder is tested by the Babcock centrifugal method after a preliminary digestion with dilute sulphuric acid, have been compared with the Roese-Gottlieb (Mojonnier) method. From the results of these comparisons which are shown in table 9, it will be noted that the Redmond method does not compare in accuracy with the Roese-Gottlieb (Mojonnier) method. It would seem that this method can only be used as a rough control

TABLE 9

Roese-Gottlieb (Mojonnier) and Redmond methods compared.

SAMPLE NUMBER	REDMOND METHOD	ROESE-GOTTLIEB (MOJONNIER) METHOD
	<i>per cent fat</i>	<i>per cent fat</i>
1	33.30	28.77
2	28.08	28.55
3	28.08	27.40
4	28.08	27.24
5	27.36	26.85
6	56.88	55.41
7	55.62	54.02
8	56.39	54.08
9	55.98	52.81

method and even under these circumstances it appears to be of little value for close checking purposes. It is believed that the multiplication factor which must be used to obtain the final result is largely responsible for the discrepancies. The Redmond method results shown in table 9 are the averages of duplicate determinations. The maximum variation between duplicates of any one sample was 1.8 per cent.

SUMMARY

A review of the results recorded herein seem to warrant the following conclusions:

1. The normal variations in results from the same sample of milk powder analyzed by the Roesse-Gottlieb (Mojonnier) method should not be over 0.15 per cent when results are expressed on the original powder basis. If results are expressed on the moisture-free basis, the variation between duplicate samples may be increased due to discrepancies in the moisture determinations.

2. There is no positive evidence of less fat being extracted from old milk powder than fresh powder when both samples are expressed on the moisture-free basis. If results are expressed on the original powder basis, there is an apparent decrease in fat content of old powders due to the fact that they may have absorbed moisture.

3. The simple ether extraction method gives results from the double roller process powder which are on an average about 0.25 per cent lower than the Roesse-Gottlieb (Mojonnier) method. Duplicate results from the former method are also subject to wider variations than those from the latter method. From sixty-nine comparisons of duplicate determinations by the Roesse-Gottlieb (Mojonnier) method there is an average variation of 0.147 per cent; a maximum of 0.49 per cent and a minimum of zero. From the ether extraction method from twenty comparisons there is an average variation of 0.369 per cent; a maximum of 1.29 per cent; and a minimum of .05 per cent.

4. On the basis of results presented herein, the Redmond modification of the Babcock centrifugal method is very unreliable. The wide variations between the two methods is undoubtedly accounted for to some extent by the large multiplication factor required for converting the test bottle reading to percentage fat in the powder.

5. The modified Roesse-Gottlieb procedure from which extraction is made from an acid medium instead of an alkaline medium gave higher results in the majority of cases than did the regular extraction from an alkaline medium. The full significance of these higher results cannot be stated however, without further investigation as to the variability of numerous results from the same sample of powder.

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IS LIPASE A NORMAL CONSTITUENT OF COW'S MILK?¹

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The evidence that a true lipase which accelerates the hydrolysis of milk fat is a normal constituent of cow's milk is not only inadequate but contradictory as well. It has been customary for most writers in reviewing this question to cite the experiments of Spolverini (1, 2), Marfan and Gillet (3), Moro (4), Gillet (5), Friedjung and Hecht (6) and Hippus (7) as having demonstrated the presence of lipase in milk, and possibly refer to the experiments of Vandeveld (8) who failed to find this enzyme. As a matter of fact, the examination of the original reports of these various investigators reveals the following facts:

Spolverini (1) states that he demonstrated lipase to be more or less active in the milk of the cow, goat, ass, and dog and also in human milk. Neither in this paper nor in the abstract of a later paper (2), where the same statement is made, is any experimental evidence whatever presented. The writer is of the opinion that Spolverini applied a test for esterase, previously worked out by Marfan and Gillet for human milk, to the milk of the animals stated.

Marfan and Gillet (3) added 10 cc. of 1 per cent monobutyrin solution and a few drops of alcoholic phenolphthalein to 1 cc. of cow's milk and after incubating for 20 minutes at 25°C. added sodium carbonate solution (approximately 0.5N) to the milk until it was neutral to the indicator. The lipase activity of the milk was reported as the number of drops of Na₂ CO₃ solution required. This is stated to be 6 to 8 drops for cow's milk as compared to 20 to 30 for human milk under identical conditions. No antiseptics are mentioned as having been employed nor were

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any controls used. The conclusion is drawn by these authors that the presence of a monobutyrylase in cow's milk, which they believe they demonstrated, is not, however, an index of the presence of a true lipase. This statement of Marfan and Gillet seems to have been overlooked by most writers.

Moro (4) attempted to ascertain whether milk contains a true lipase which hydrolyzes neutral fat, but his experiments were with human milk only. The method was qualitative, and was rather novel, although its accuracy might be questioned. A small amount of neutral olive oil was added to the fresh milk in a small test tube and the mixture shaken gently, but not hard enough to cause a permanent emulsion. The tube was then incubated at 38°C. for a given period and the oil floating on the surface transferred to a 0.25 per cent sodium carbonate solution on a watch glass. If an emulsion occurred it was concluded that free fatty acids were present and the milk had contained a true lipase. According to Moro positive results were secured by applying this test to human milk.

Gillet (5) applied the method of Marfan and Gillet (3) again to cow's, goat's and ass's milk and also to human milk. His results, expressed in terms of drops of 0.5N Na_2CO_3 solution to neutralize the acid liberated from 10 cc. of 1 per cent monobutyryl solution by 1 cc. of milk after twenty minutes incubation at 25°C., were as follows: Human milk, 20 to 30 drops; cow's milk, 6 drops; ass's milk, 9 drops; goat's milk 2 drops.

Friedjung and Hecht (6) modified the method of Marfan and Gillet slightly, but applied it to human milk only. They concluded from their results that the evidence is conclusive that a monobutyrylase is present in human milk. The statement is made, however, that they did not attempt to ascertain whether a ferment is present which splits milk fat, also.

Hippius (7) attempted to demonstrate true lipase in human milk using a method similar to that employed by Moro, except that the free fatty acids liberated from the oil which was added to the milk were detected by means of an indicator. Positive results were secured by this method. No tests were made on cow's milk.

Vandeveld (8) carried out some careful experiments on the possibility of a true lipase being present in cow's milk. He rightly expressed the opinion that a study of the decomposition of fat in milk should be made on milk fat itself. He added 3.3 cc. of 3 per cent acetone solution of iodoform as antiseptic to 25 cc. of cow's milk in several tests from individual cows, and determined the increase in acidity after fifty-nine days in the incubator at 37.5°C. Acidities were determined (a) by steam distillation and titration of 50 cc. of the distillate after adding 50 cc. of alcohol, (b) by direct titration with 0.1N NaOH after adding 100 cc. of alcohol and 100 cc. of ether to dissolve both the fat and free fatty acids. In three experiments by the distillation method no increase whatever in acidity was noted, and identical results were secured in four experiments using the direct titration method. These results of Vandeveld's are open to the criticism that no study was made of the effect of the antiseptic employed on lipase activity. The writer has shown recently (9) that both acetone and iodoform have a marked retarding effect on lipase activity and if the solution of the antiseptic is slightly old so that a little free iodine is present, lipase activity is completely inhibited. More valid evidence of the absence of lipase in cow's milk was obtained by Vandeveld in another single test in which raw sterile milk, containing no antiseptic, was examined for free fatty acids in the extracted fat after having stood for fifteen months. The amount of free fatty acids in 5 grams of extracted fat was negligible, the fat requiring only 2.2 cc. 0.1N alc-KOH to neutralize the free fatty acids. Vandeveld drew the conclusion from his studies that the lipolytic decompositions which have been observed in cow's milk were the results of poorly conducted studies or failure to eliminate bacterial contamination.

From the experiments of Davidsohn (10) it seems doubtful, also, that cow's milk contains a lipase which splits even a simple glyceride like tri-butyryl. This investigator states that cow's milk can be differentiated from human milk by the fact that the latter will set free butyric acid from tri-butyryl in a few minutes, while cow's milk or boiled human milk will not do so.

Another more recent experimental study of milk enzymes, which failed to demonstrate lipase in a product made from cow's milk, was that of Thatcher and Dahlberg (11). The curd of raw, fresh and stored butter was mixed with 5 per cent butter fat or olive oil, in the presence of 2.5 per cent chloroform, and the acidity while fresh and after four days' incubation, compared with that of control samples which had been boiled. No increases in acidity were noted which could be attributed to lipase activity. The experiment is open to the criticism that chloroform has a marked retarding action on lipase activity, as the writer has shown in recently published (9) studies.

Two studies which resulted in positive indications of lipase in cow's milk were carried out by Rogers (12) and later by Rogers, Berg and Davis (13). In the first study fresh cow's milk was mixed with an equal volume of heated butter fat and the initial acidity of the mixture compared with that after 13 and 19 days' incubation, using boiled fresh milk as control. An increase in acidity of 0.9 cc. of 0.1N acid per 100 cc. of milk was noted after nineteen days over the control. Formaldehyde 1:1200 was used as antiseptic. In another experiment Rogers divided fresh cream into two portions, heated one portion to 60°C. for fifteen minutes to destroy the lipase, if present, and churned both samples of cream. The butter was melted and salted in the melted condition and also treated with formaldehyde, so that the water in the butter contained 1 part in 1500 of the antiseptic. The acidity of the heated and unheated portions of butter after forty-eight and ninety-two days at 23°C. showed a small but distinct increase in the unheated portion which could be attributed to lipolytic activity.

In the experiments by Rogers, Berg and Davis, 50 cc. portions of milk which had been pasteurized at different temperatures were treated with 1 per cent CHCl_3 and 1 per cent ethyl butyrate. The flasks were neutralized at once to phenolphthalein using 0.1N NaOH and again at twenty-four hour intervals following incubation at 26°C. The results were compared with controls to which no ethyl butyrate was added. The results show that cow's milk has an appreciable ability to hydrolyze an ester like ethyl

butyrate under these conditions and that this ability is greatly weakened when the milk is heated to 66°C. (flash pasteurization) and destroyed at 80°C. In the writer's opinion these results should not be interpreted, however, as indicating the presence of a true lipase in cow's milk.

EXPERIMENTAL

In an examination of the possibility that cow's milk may, at times, undergo lipolytic fermentation which can not be attributed to bacteria or other contaminating organisms, the writer was impressed with the lack of definite data on the question whether cow's milk normally contains a true lipolytic ferment. The experiments regarding the abnormal condition referred to will be reported in another paper. In the present paper are reported the results of the study of a possible lipolytic ferment in normal milk.

Eleven experiments in all were carried out on this question. No foreign fats or single glycerides or esters were added to the samples under investigation because the problem involved solely the determination of a natural true lipase in the milk.

The technic used in the various experiments was not the same throughout the entire study. It was essentially identical for the first six experiments; it was varied materially in the seventh; and the last four experiments were performed using a still different technic. The experiments naturally fall, therefore, into three groups, altho experiment 7, alone, constitutes one group. It does not seem necessary to report each experiment in detail. The general features of each method will accordingly be described together with a summary of the results and the reasons for modifying the technic.

It may be mentioned that formaldehyde in the proportion of 1 part HCHO to 1500 to 2000 parts of milk, was used in all cases. The use of formaldehyde was based on experiments which have been reported elsewhere (9), showing that this antiseptic in this concentration has no detrimental effect whatever on the activity of animal lipase.

The source of the milk in all experiments was fresh milk from cows in the University herd. The milk in no case was more than a few hours old when the experiments were begun.

Experiments 1 to 6

In these experiments the technic employed was adapted from that used by Rogers, Berg and Davis (13) except that no ethyl butyrate was added. In detail, the method was essentially as follows in each experiment, each test being run in duplicate.

Method. Fifty cubic centimeter portions of milk and 25 cc. of sterile distilled water were introduced into sterile 200 cc. Erlenmeyer flasks. One cubic centimeter of neutral, saturated potassium oxalate solution was added to each flask to reduce the initial titration value and also to increase the sharpness of the end point, and the mixture then allowed to stand at least two minutes. Formaldehyde was next added to each flask in amount so that the concentration would be 1:1500 to 1:2000 after the initial titration had been made. Five drops of 1 per cent alcoholic phenolphthalein solution were then added and each flask titrated at once to a two-minute pink with 0.1N alkali (aqueous). All flasks were incubated at 37°C. with frequent shaking, the flasks being stoppered with cotton plugs. The titrations were repeated at twenty-four-hour intervals for from two to five days in the various experiments. Each experiment included flasks containing (a) raw whole milk, (b) raw skim milk (prepared from the same milk by centrifugal separation), (c) raw whole milk heated to 80°C. for one minute, (d) raw skim milk heated to 80°C. for one minute. Either the heated skim milk or whole milk flasks were omitted in experiments 4, 5 and 6, one heated control being regarded as sufficient after it was found that there was no difference between the acidity which developed in the whole and skim milk samples after heating. In experiments 4 and 5 the whole milk was prepared by adding the cream from the centrifugal separation to portions of the skim milk.

The results of the experiments using this technic are summarized in table 1. The titrations have been calculated to cubic centimeters of 0.1N acid developed in 100 cc. of milk, which

could be attributed to lipolytic activity. In calculating the data the titration values obtained for the heated whole milks are subtracted from the titration values of the raw whole milks leaving the net acidities which could have developed in the whole milk as the result of lipase and other enzyme actions. Similarly, the difference between the acidities of the raw and heated skim milks gives the net acidities which developed in the milk plasma due to enzymes other than lipase. Where only one heated sample was run the result was used for both the whole and skim milk calculations. The assumption that the skim milks were free from fat was based on the fact that no demonstrable amount of

TABLE 1

Lipase activity of cow's milk as measured by direct titration at intervals following incubation

EXPERIMENT NUMBER	NET ACIDITY* IN WHOLE MILK AFTER			NET ACIDITY* IN SKIM MILK AFTER			ACIDITY* DUE TO LIPASE ACTIVITY AFTER		
	1 day	2 days	5 days	1 day	2 days	5 days	1 day	2 days	5 days
	cc.	cc.	cc.	cc.	cc.	cc.	cc.	cc.	cc.
1	0.90	1.14	2.14	0.64	0.94	2.88	+0.26	+0.20	-0.74
2	1.96	1.60	2.35	2.06	2.76	3.64	-0.10	-1.16	-1.29
3	0.82	0.98	1.30	0.70	0.20	1.30	+0.12	+0.78	0.00
4	1.50	1.80	2.04†	1.86	3.00	3.24†	-0.54	-1.20	-0.96†
5	1.50	2.54	3.10†	1.34	1.74	2.30†	+0.16	+0.80	+0.80
6		1.00			0.80			+0.20	
7	2.72	2.10	2.60†	2.00	1.20	1.10	+0.72	+0.90	+1.60

* Cubic centimeters of 0.1N acid in 100 cc. milk.

† Three days in the case of experiments 4, 5, and 7.

fat was left in the skim milk after it had been passed through the centrifugal separator twice at high speed and a temperature of 95°F. The difference between the net acidities in the whole and skim milks gives the true acidity due to the action of lipase on the milk fat.

An inspection of table 1 shows very clearly that the presence of a true lipase in cow's milk is not demonstrated by the method employed in experiments 1 to 6. Whether these results are to be accepted without qualification, or not, will be brought out more clearly in the ensuing discussion and in the report of the other experiments.

The interesting feature of these experiments was the fact that acidities developed from day to day in the control samples, i.e., those which had been heated as well as in those which had not been heated. In order to throw some light on the cause of this phenomenon, some tests were made in imitation of the lipase experiments using, in one case, a solution of salts² which were intended to be a duplicate of the mineral salts of milk, both qualitatively and quantitatively, and using, in another case, serum from whole milk, prepared by filtering formaldehyde treated milk through a Pasteur-Chamberland filter under pressure. The following flasks were prepared and treated as in the case of the lipase studies:

Flask 1. 50 cc. salt solution + 25 cc. water + 0.5 cc. saturated potassium oxalate solution.

Flask 2. 50 cc. salt solution + 25 cc. water.

Flask 3. 50 cc. milk serum + 25 cc. water + 1 cc. saturated potassium oxalate solution.

Flask 4. 50 cc. milk serum + 25 cc. water.

The initial acidities and the acidities which developed on incubation are shown in table 2, the results being calculated in terms of cubic centimeters of 0.1N acid per 100 cc. The data in this table are not directly comparable with those given in table 1 where only the net acidities developed in the samples of milk are reported. It may be stated, however, that in general, the values given for the acidities developed in flasks 2 and 4, table 2, are about one-half of those obtained in the experiments using milk. A consideration of the data in table 2, together with this

² The salt mixture was that given for cow's milk by Van Slyke, L. L., and Bosworth, A. W., (N. Y. Agr. Exp. Sta. Tech. Bul. 39, 1914), and consisted of the following salts per liter of milk.

2.212 grams $\text{CaHPO}_4 \cdot 2\text{H}_2\text{O}$
 1.190 grams CaCl_2
 1.080 grams $\text{MgH}_2(\text{PO}_4)_2$
 3.040 grams $\text{NaC}_6\text{H}_7\text{O}_7 \cdot \text{H}_2\text{O}$
 0.540 grams $\text{KC}_6\text{H}_7\text{O}_7 \cdot \text{H}_2\text{O}$

The CaHPO_4 was merely suspended in the solution, care being taken to mix it thoroughly before withdrawing portions for the tests.

fact, shows very clearly that the acidity which apparently develops from day to day when sterile milk is incubated is due in large part to a reversion of some of the salts formed in titration. Apparently this phenomenon can be prevented in part by precipitation of the calcium salts as calcium oxalate. This suggests that a part of the reversion to acid salts is due to the hydrolysis of CaHPO_4 , which is known to occur, especially in the presence of alkali. This source of titratable acidity can be removed by oxalate. In all probability the MgHPO_4 , which forms in the titration of $\text{MgH}_2(\text{PO}_4)_2$ hydrolyzes in the same manner. Magnesium salts, however, can not be removed by addition of oxalate.

TABLE 2

Acidity developed in milk salt solution and in milk serum on incubation

FLASK	CHARACTER OF MIXTURE	INITIAL TITRATION	ACIDITY DEVELOPED IN		
			1 day	3 days	5 days
		cc.	cc.	cc.	cc.
1	Salt solution, oxalated.....	8.90	1.40		2.94
2	Salt solution, untreated.....	11.80	1.80	3.80	5.50
3	Serum, oxalated.....	8.04	0.44	1.84	2.24
4	Serum, untreated.....	11.10	0.96	1.76	4.10

Experiment 7

The results secured in experiments 1 to 6, together with those obtained in the supplementary experiment on the cause of the acidities which developed from day to day in the control samples, prompted a careful consideration of the technic employed. This study led to the conclusion that the following serious criticisms could well be directed against this technic.

1. Acidity develops in both the experimental and control samples which is not due to enzyme action, and may vary with the amount of alkali added.

2. Neutralization of milk to a visible, satisfactory end point, using phenolphthalein as indicator, means that the relation of the milk is considerably on the alkaline side of neutrality, being at $\text{pH} = 9$ or over. This means that whatever lipase is present

in milk is being allowed to act in a hydrogen ion concentration considerably on the alkaline side of the normal hydrogen ion concentration of milk. Altho it is not known what effect this would have on the activity of lipase in milk, if present, the possibility of it being affected one way or the other is sufficient grounds for criticism.

3. If lipase is a normal constituent of milk, there is no basis for supposing that it exerts a selective action on the glycerides, liberating only those fatty acids which are soluble in an aqueous medium. Obviously the technic should be modified to permit the titration of both the water soluble and the water insoluble fatty acids which are liberated.

With these points in view another experiment was conducted in which the following technic was used.

Duplicate 250 cc. portions of milk were treated with formaldehyde 1:2000 and the initial acidity of each portion determined on a 50 cc. aliquot withdrawn with a pipette. The remainder of the milk was transferred to sterile 300 cc. Erlenmeyer flasks, stoppered with cotton plugs and the acidity determined on 50 cc. aliquots after incubation for twenty-four, forty-eight and seventy-two hours at 37°C. The acidity was determined in each case by adding 1 cc. of neutral, saturated potassium oxalate to the 50 cc. portion of milk, letting stand for several minutes, adding 250 cc. of neutral acetone-alcohol (1:1), filtering off the precipitate, after vigorous shaking, washing the filtrate several times with neutral acetone-alcohol (1:1), and titrating the combined filtrate and washings with 0.1N alcoholic KOH. In this experiment rosolic acid was used as indicator, but it was not found to have any advantages over phenolphthalein notwithstanding the fact that it changes color at $\text{pH} = 7$.

The results of this experiment, using the above technic, are shown in table 1, experiment 7, the data being calculated to values corresponding to the other experiments given in this table, taking into account the fact that the titrations of the aliquots after the twenty-four, forty-eight and seventy-two-hour incubations included the initial titratable acidity of the milk.

The data show that while the results in the last three columns indicate a slight development of acidity which could be attributed to lipase activity, at the same time it is apparent that this was due to an actual decline in the net acidity of the skim milk, and not to an actual increase in the net acidity in the whole milk samples. These data, therefore, like the data from experiments 1 to 6, do not indicate the presence of lipase in normal milk.

Experiments 8 to 11

In these experiments a somewhat different technic was followed in that in place of using boiled or heated milk as control samples, an antiseptic was added to the raw milk which had been found

TABLE 3

Lipase activity of normal milk indicated by comparison with milk to which lipase paralyzoz had been added

EXPERIMENT NUMBER	ACIDITY* DEVELOPED IN MILK IN		ACIDITY* DEVELOPED IN CONTROL IN		ACIDITY* DUE TO LIPASE IN	
	24 hours	48 hours	24 hours	48 hours	24 hours	48 hours
	cc.	cc.	cc.	cc.	cc.	cc.
8	3.74	2.30	-0.04	3.04	3.78	-0.74
9	-0.60		1.80		-1.20	
10(a)		0.60		-2.87		0.60
10(b)		2.44		-1.18		2.44
10(c)		3.90		-0.36		3.90
11		-0.56		2.96		

* Cubic centimeters of 0.1N acid in 100 cc. milk.

by experiment (9) to completely inhibit lipase activity. The antiseptic used for this purpose was iodine dissolved in KI. The skim milk controls were omitted, also, although the wisdom of this may be questioned inasmuch as results were secured in certain supplementary experiments in which a proteolytic enzyme was added to milk containing iodine, which indicated that proteolysis is not inhibited completely by this antiseptic.

Formaldehyde was used as bacterial antiseptic as in the other experiments and acidities were determined on fresh samples and after incubation as in the case of experiment 7. The technic

of this operation was modified to advantage in these experiments in the following manner. Saturated neutral potassium oxalate was added to the aliquot withdrawn for titration and after standing a few minutes was titrated to a pink color with 0.1N alcoholic KOH, using phenolphthalein as indicator. Two hundred cubic centimeters of alcohol-ether (1:1) were now added and the titration of the dissolved fatty acids completed. All tests were run in duplicate.

The milk used in experiment 8 was mixed milk from the University herd. In experiment 9 the mixed milk from two cows, only, a Jersey and a Holstein, was used. In experiment 10 the milk from each of three individual cows which had been in milk 411, 429 and 390 days, respectively, was tested for lipase by this method. Experiment 11 was a repetition of the work on the milk of cow 502, tested in experiment 10, the retest being made thirty days later.

In calculating the results of these experiments in terms of possible lipase activity only the increase in acidity of the milk containing added iodine was subtracted from the increase in acidity in the milk to which no lipase paralyzator was added. The amount of iodine solution added varied a little in the different experiments, the idea being to furnish a surplus of free iodine after the iodine had reacted with all the casein in the milk. In experiment 8 the amount added was 8 cc. of 5 per cent solution in 5 per cent KI to 90 cc. of milk. In Experiments 9, 10, and 11 this was 6 cc. of 10 per cent iodine in 12 per cent KI.

The results of the experiments are summarized in table 3. Only in the case of Experiment 10 was any definite indication of lipase activity suggested. This was not confirmed in Experiment 11 using milk from the same cow a month later.

SUMMARY

Experiments are reported using several different methods of analysis which fail to show that cow's milk normally contains active lipolytic ferment.

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PROOF OF THE PRESENCE OF LIPASE IN MILK AND A NEW METHOD FOR THE DETECTION OF THE ENZYME

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There are now recognized several distinct enzymes which induce hydrolysis of fats and the simpler esters. These are grouped as lipolytic enzymes. The various members of the group are distinguished first according to their origin, for instance, the lipolytic enzyme of the stomach differs in properties from that of the pancreas (1) also, those from plants react differently from those of animal origin (2). Secondly, these enzymes are distinguished by the ester which each attacks most easily. Monobutyrylase is the name given to the enzyme which splits monobutyryl (3). The term esterase is used for that subgroup of lipolytic enzymes which split the simple esters more easily than the natural fats (4). The various members of this group may act on more than one ester but it is usually found that one ester is hydrolyzed more easily than another (5).

The term lipase has in late years been used to describe those enzymes which hydrolyze the natural glycerol fats. They may also split the esters of the lower alcohols and acids, but they must act on the true fats to be known as lipases (4).

I. PREVIOUS METHODS FOR THE DETECTION AND DETERMINATION OF LIPASE

In most of the methods in use a convenient ester is chosen as substrate and after adding the extract to be tested, the mixture is titrated at the beginning and at the end of a period of time. The increase in acidity, which is due to the splitting off of acids by the enzyme, is taken as a measure of the lipolytic activity. When the digestion period is short no preservative is used, but if the time and temperature are such that the reaction is likely to be complicated by bacterial growth, then there is added a preservative to check such action.

1. The substrate

On account of the easy solubility of the lower esters, they have been most frequently used as substrates to test the action of lipase. Objection has been raised to this practice (6) since by definition lipolytic enzymes other than lipases may hydrolyze these. The difficulty in using the natural fats is in the fact that they are immiscible with water. The enzyme seems not to dissolve in the fat (7) but remains in the water layer, it can, therefore, act only on the fat at the interface. The action on the fat cannot be rapid unless it is emulsified in the water phase thus exposing a large surface of fat to the action of the enzyme (8). There has been much difficulty in preparing emulsions of the natural fats of such stability and uniformity that comparable results can be obtained at different times and on different enzyme extracts. In order to secure the desired emulsification of the fat there have been employed gums such as gum arabic, and also bile salts (9), but in some experiments dependence is placed merely on frequent shaking (10).

Aside from the simple esters of the lower alcohols and acids a great variety of which have been used as substrates (11), the following fats and oils have been employed rather commonly to determine lipolytic activity; cottonseed oil (12), castor oil (12), egg yolk (13), lard (10), cod liver oil (14), butter fat (15), almond oil (16), lecithin (17), olive oil (18), and linseed oil (19).

2. Methods of following activity of the enzyme on the substrate

As has already been mentioned the great majority of investigators make determinations by titration of the acid liberated from the esters and take these results as a measure of lipolytic activity. If aqueous alkali is used in this titration the results are likely to be too low when natural fats are employed on account of the fact that some of the fatty acids are insoluble in water and may not therefore react with the alkali. To lessen this error it has been recommended that alcohol be added to the solution to be titrated (20). This refinement has been frequently omitted by investigators, however.

Rachford (21) describes a method for determining lipolytic activity by using olive oil as substrate, and measuring the time necessary for sufficient fatty acid to be produced to permit easy emulsification of the mixture when brought in contact with a solution of sodium bicarbonate.

Where any salicylic acid ester is used as substrate, the salicylic acid liberated can be determined colorimetrically by use of ferric chloride (22).

If an ester is employed as substrate which splits into products that greatly alter the surface tension of the solution, then surface tension determinations can be made use of to follow lipolysis (23). Determinations of hydrogen-ion concentration (24), and of electrical conductivity (25) have also been suggested as means of following the splitting of fats by enzymes.

A unique method for detecting lipase was suggested by Carnot and Mauban (26), the fat is fixed in a solid media of agar and after the enzyme has been allowed to act at the surface, a color reaction with copper sulphate indicates that lipolysis has taken place.

3. The use of preservatives in lipolytic digestion experiments

Reactions brought about by enzymes and by bacteria are strikingly similar (27). Where enzyme digestions are proceeding in solutions suitable for bacterial growth the measurement of enzyme activity may be complicated by changes brought about by bacteria. Usually the substrate or the enzyme extract contains substances which bacteria break down; the temperature favorable for enzyme action is also very favorable for microorganisms. Commonly, preservatives are added which it is intended will not in the least check enzyme action but which will entirely prevent bacterial growth. A preservative which fulfills these requirements is not known, though there are a few that approximate the ideal.

In studies of enzyme lipolysis one of the first preservatives to be used was chloral hydrate (28) but it has been shown that this inhibits to some extent the action of lipase (29). Chloro-

form has been objected to on the same ground (30), though it has been used as an antiseptic frequently and has been particularly recommended for use in experimental work on milk enzymes (31); however, Vincent (32) found chloroform quite ineffective in preventing lactic fermentation. Kastle and Loevenhart (33) made an extensive study of the effect of common antiseptics on lipase, and found the greater number to be harmful.

It is significant that Vandeveld who is most frequently quoted in support of the hypothesis that milk contains no lipase (34) used iodoform in acetone as antiseptic. Yet it has been shown by Palmer (35) that lipolytic activity is nearly prevented by the presence of these substances. Toluol is probably the most commonly used antiseptic though formaldehyde has been found to have very little, if any, retarding influence on lipolytic hydrolysis (36).

In many of our preliminary experiments in which milk or butter fat was the substrate, bacterial action was not prevented by the liberal addition of toluol, chloroform, thymol or formaldehyde. In testing extracts high in lipase, and by use of such substrates as the simple esters, hydrolysis takes place so rapidly that there can be no confusion on account of bacterial action. However, such substances as milk which contain but small amounts of enzyme and where the natural fats are employed as substrates the hydrolysis proceeds so slowly and such a long digestion time is necessary (usually several days) that a thoroughly efficient antiseptic must be used. All media containing milk constituents are especially favorable for the activity of microorganisms.

Unfortunately much work has been done upon which reliance cannot be placed on account of the choice of preservative. Either it has been too weak to check bacterial growth or it has been of such nature that enzyme action is also inhibited.

Vandeveld (37) believed that cow's milk does not contain lipase and that the decomposition which others observed was due to improper control and especially to the presence of bacteria.

Saxl (38) concluded that none of the previously recommended methods affords a quantitative study of the ester splitting, since

the values obtained either are so slight on the short periods of digestion that they scarcely lie beyond the limits of error, or, on longer periods of digestion are obscured by the sources of error which are connected with the methods of determination.

Thatcher and Dahlberg (39) speaking particularly of lipase point out the "confusion which exists with reference to the normal enzymes of milk. The contradictory evidence which has been presented undoubtedly results in part, at least, from the rather crude methods of study of enzyme action which were in use at the time when those investigations were in progress."

II. PREVIOUS EXPERIMENTAL WORK ON THE PRESENCE OF LIPOLYTIC ENZYMES IN MILK AND MILK PRODUCTS

Marfan in 1901 (40) was probably the first to report that experimental proof had been obtained of the presence of a lipolytic enzyme which splits monobutyryl in the milk of woman and the cow. Detailed results were not published however until later (41). In these experiments monobutyryl was added to the milk and after a digestion period the acidity was titrated. It was assumed that any increase in acidity was due to butyric acid split off from the ester. The results can quite properly be objected to on the ground that no antiseptic was used to prevent bacterial growth; however the weight of evidence was greatly in favor of the conclusions reached.

Luzzati and Biolchini (42) and Spolverini (43) used the same substrate as Marfan and Gillet and followed a similar procedure except that after the digestion period the acid was distilled and the distillate titrated. They assumed that no volatile acid could be produced in the digestion mixture in any other way, even by bacteria, than from the action of lipolytic enzyme on the substrate setting free volatile butyric acid.

It will be noted that in the investigations just described the substrate used was always monobutyryl. Since it is understood in this work that lipase must split neutral natural fats, the presence of such an enzyme in milk was not necessarily proved. The methods which were used would indicate only one of the esterases—monobutyrylase.

Moro (44), on the other hand, employed olive oil as substrate and using the method of Rachford (21) found that milk was able to induce hydrolysis. This was the first time that a true fat-splitting lipase was identified in milk.

Friedjung and Hecht (45) demonstrated splitting of monobutyrin by milk. They used thymol as preservative and titrated both the digestion mixture and the distillate from same.

Gillet (46) found monobutyrylase in milk; he used as an antiseptic sodium fluoride and chloroform and titrated the acid split off directly without distillation. He obtained no enzymic splitting of triacetin or of a neutral fat substrate prepared from neat's foot oil, hence he concluded that no true lipase was present in milk.

Rogers (47) by preparing butter from fresh unheated cream and also by mixing raw milk with butter and adding formaldehyde as a preservative observed considerable increase in acidity which was not found in the heated check runs. He believed that this was due to the presence of natural lipase in the milk which hydrolyzed butter fat.

Hippius (48) using almond oil as substrate determined the lipolytic activity of woman's milk which had been heated to various temperatures. He made no attempt to estimate quantitatively the lipolytic activity but by use of an indicator particularly sensitive to fatty acids he determined whether or not the neutral oil had been split.

Koning (49) did not find the percentage of fat in milk to decrease on standing with an antiseptic, and concluded, therefore, that no lipase was present.

Vandeveldt (37) using the fat of milk as substrate and iodoform in acetone as antiseptic allowed digestion to proceed for several weeks at incubator temperature. The acidity of the mixture and also of the distillate was determined at the beginning and at the end of the period. Since no increase in acidity was found he believed that no lipase was present.

Vincent (50) made a few determinations and was not able to prove the presence of fat-splitting enzyme in milk or cream.

Grimmer (51) found a monobutyrylase along with other enzymes in extracts made from the milk glands of the cow.

Davidsohn (24) used tributyrin as substrate and changes in surface tension as a measure of the splitting. There was added to the digestion media also disodium phosphate in order to preserve a reaction which he considered most suitable for the enzyme. He found very little lipolytic power in cow's milk but much more in woman's milk. He was unable to find a satisfactory preservative for use in the digestions and he admitted on this account that there was some weakness in his evidence, but believed notwithstanding this point that his conclusions were entirely warranted.

Thatcher and Dahlberg (39) used butter and olive oil as substrates and chloroform as antiseptic. They concluded that "lipases are present in butter in very small amounts, if at all."

Resch (52) followed methods similar to those used by Davidsohn but tested woman's milk only. The presence of lipolytic enzyme was found in all stages of lactation.

Palmer (53) has reported some investigations on the lipolytic activity of milk. His substrate consisted of an artificial milk prepared by emulsifying butter fat in water with gum arabic. Formaldehyde was used as preservative; acidity was determined before and after digestion by titration in presence of acetone and ether with alcoholic KOH. He states that "the work has not progressed to the point where it can be stated with assurance whether or not lipase is a normal constituent of milk."

Thus, it is seen that the question of the presence of lipase in milk has not been answered. While some investigators believe that they have established proof of an enzyme which will split neutral natural fat, others have been equally certain that milk has no such property, and many have been undecided even after careful experimentation. In most cases the methods used in testing for lipase are not free from objection, if no preservative has been added there is the possibility of a reaction taking place in the digestion media due to bacteria and not distinguishable from the enzymic reaction; on the other hand, if a preservative has been used there are the two possibilities that it may not have

been sufficiently powerful to check bacterial growth, or, it may have been so active as to inhibit the action of the enzyme also.

These remarks are to be understood to apply to true lipase. There has been an agreement with respect to monobutyrinase, no investigator having failed to find this enzyme.

In this connection should be mentioned the work of Pastrovich and Ulzer (54) who showed that fats were split under some conditions by proteins; casein was among those found to produce this effect. The theory that milk fat is split through the chemical action of protein was considered more or less favorably by Raudnitz (55). Falk (56) found a relation between lipolytic activity and chemical structure. However a careful inspection of the findings of all investigators on lipolytic action would indicate that in the large majority of cases, at least, the reaction is induced by a typical enzyme, rarely could the hydrolysis of the esters be attributed to chemical substances.

III. A NEW METHOD FOR THE DETECTION AND ESTIMATION OF LIPASE

From the discussion in the first part of this paper it is seen that the requirements of a dependable method for the detection of lipase are, (1) that a natural fat be employed as substrate, (2) that this fat be well emulsified in the aqueous solution, and (3) that a preservative be used which prevents the growth of organisms but does not inhibit lipolytic action.

Such an ideal method is very nearly reached in the following procedure: Cream of high fat (40 to 50 per cent) is employed. To this is added cane sugar in sufficient quantity to produce a saturated solution with the water present and with any water which may be later added in the enzyme extract. In order to bring this about, two parts of sugar should be added to one part of water.

The cream-sugar mixture is boiled to facilitate solution of the sugar and to destroy enzymes of the cream. Upon cooling, the milk or enzyme solution to be tested is added and the whole incubated at 38° to 40°C. for a considerable period of time. This may vary from three to thirty days or longer depending on the amount of enzyme present.

Acidity is determined at the beginning and end of the digestion period as follows: Ten grams of the sample is diluted with 50 cc. of distilled water. Neutral phenolphthalein is added and the mixture titrated to neutrality using 0.1N NaOH.

If the quantity of enzyme present is considerable, or, if the digestion period is sufficiently long, the characteristic odor of free butyric acid becomes evident.

The advantages of this method are that a neutral natural fat is used as substrate which is well emulsified in the water. The sugar increases the viscosity of the mixture to such an extent that there is no separation of fat, except after a very long time. However if such does take place, the mixture can be easily made homogeneous again by stirring. The sugar prevents bacterial growth but does not injure the enzyme nor prevent its action.

Although the effect of saturated sugar on other enzymes has not been investigated it would seem that the procedure applied here in the study of lipolytic enzymes might also find application in the study of many other enzymes where preservatives are necessary.

Experiment 1

The following is an example of the use of this method on the lipase of commercial pancreatin: One-tenth per cent of the powder was made up with water and then added to the cream-sugar substrate. The quantities of 0.1N NaOH necessary per 100 grams of the digestion media at various intervals are as follows: Beginning—4.0; three days—6.5; 7—8.5; 9—13.5; 13—22.0; 16—23.5; 23—26.5; 29—41.0; 77—86.5.

An accompanying check was run wherein the enzyme infusion was boiled. No increase in acidity was noted.

During the latter part of the digestion period a strong odor of butyric acid was evident. Bacterial count at the beginning of the digestion period showed 460 per gram, and at the end of twenty-nine days—90 per gram. In this determination room temperature was used for digestion and no effort was made to hold it uniform from day to day. This will account for the irregularity of acidity production at different intervals during the digestion period.

IV. THE DETECTION AND ESTIMATION OF LIPASE IN MILK

Experiment 2

To the regular cream-sugar substrate was added raw milk in different proportions. The temperature was again that of the room. The numbers of cubic centimeters of 0.1N NaOH required per 100 cc. of the digestion mixtures are here recorded.

TIME	PROPORTION OF RAW MILK ADDED					
	1 per cent	3 per cent	5 per cent	10 per cent	15 per cent	20 per cent
	cc.	cc.	cc.	cc.	cc.	cc.
Beginning.....	3.5	3.5	4.0	4.0	4.5	4.5
8 days.....	4.5	5.5	10.0	12.0	13.0	16.0
16 days.....	6.5	10.0	11.0	14.5	15.5	20.0
22 days.....	6.5	10.0	12.0	14.5	15.5	24.5
29 days.....	6.5	10.0	14.5	18.5	17.5	28.5
36 days.....	6.5	10.0	13.5	16.5	20.0	22.0
43 days.....	6.5	9.0	12.0	15.5	19.0	25.0

During the latter part of the digestion period the odor of butyric acid was very strong in those samples containing the larger proportions of milk.

Raw milk is seen to be very active in the production of acid in the cream substrate even at room temperatures.

In the next experiment the samples were incubated at 38° to 40°C. under which conditions the enzyme is found to be much more active.

Experiment 3

Five per cent of raw milk was added to the regularly prepared cream-sugar substrate, and a check was run with an equal proportion of the same milk boiled.

TIME OF DIGESTION	0.1N NaOH PER 100 GRAMS OF MIXTURE	
	Raw milk	Boiled milk
	cc.	cc.
Beginning.....	5.0	5.0
8 days.....	14.5	5.0
14 days.....	20.1	5.5
20 days.....	22.0	5.5

The slight increase in acidity in the boiled check is due either to experimental error in reading the end point, or, to some change brought about by the action of the heat of the incubator on some of the constituents of the digestion medium.

At the end of the twenty-day period, the bacterial count on the raw milk mixture was 30 per gram. The bacteria present in this case and also in the case of the pancreatin digestion made in experiment 1 were doubtless inactive spore forms. For this reason and on account of further proof which is to follow it would not seem possible that they could have anything to do with the acid production, in which case it can only be concluded that there is an enzyme in the milk which is the cause of the change.

In order to show that the acidity produced in these digestions is due to a splitting of the milk fat the following experiment was run.

Experiment 4

A preparation was made by dissolving in skim milk sufficient cane sugar to make a saturated solution. This was divided and to each of two portions 5 per cent raw skim milk and 5 per cent of boiled skim milk were added. At the end of six weeks there was no increase in acidity. The same skim milk was found however to cause acid production when added to the cream-sugar substrate. These results show that it is only the milk fat which is acted upon by the active agency of the raw milk.

V. PROOF THAT THE INCREASE IN ACIDITY IS DUE TO ENZYMES AND NOT TO BACTERIA

The evidence thus far submitted points to the assumption that the active constituent of the raw milk is natural enzyme and that bacterial action has nothing to do with the acid production. The following points have already been offered in support of this conclusion.

1. The bacterial counts in all mixtures have been low. Especially should be noted experiment 1, where the bacterial count decreased as the acid production went on.

2. The rate of acid production increases regularly with the amount of raw milk added. If it were merely a case of inoculating the digestion media with bacteria then there would be much less difference where different amounts of milk were used.

3. The similarity of the action of raw milk to the lipase of commercial pancreatin is seen by comparing experiments 1 and 2. In addition to these experiments there have been run digestions by pancreatin and milk side by side. The action in these cases has been similar and particularly have they been found to behave alike when the acidity or alkalinity of the digestion media has been changed. There is every indication that milk lipase is identical with pancreatic lipase.

Rahn (57) in a very ingenious piece of work has shown how reactions brought about by enzymes differ in rate from those caused by bacteria. The reaction induced by enzymes is monomolecular, the greatest rate being found at the beginning and then gradually becoming less. On the other hand, when a reaction is brought about by bacteria there will be little action at first but as the numbers increase the rate increases. Of course later when the products of the reaction begin to accumulate the action again declines.

Thus, it is possible to ascertain if a given reaction is due to bacteria or to enzymes by determining the time at which the highest rate takes place. If it is at the beginning of the reaction it is certainly not due to bacteria. Bradley (58) has made measurements of the activity of pancreatic lipase, and has shown that a typical enzyme curve results.

Experiment 5

In this experiment 20 per cent of milk taken directly from the cow was added to the regular cream-sugar substrate; the temperature was carefully controlled at 40° and the acidity determined.

Following is the number of cubic centimeters of 0.1N NaOH required per 100 cc. of mixture at the beginning and each day for seventeen days thereafter:

Beginning—6.0; 1 day—9.0; 2 days—11.5; 3—14.0; 4—16.0; 5—17.0; 6—18.0; 7—19.0; 8—20.0; 9—22.0; 10—22.5; 11—24.5; 12—24.5; 13—25.0; 14—26.5; 15—27.5; 16—28.0; 17—30.0.

It is seen that the greatest activity is at the beginning of the digestion period with a slower rate thereafter. Plotting a curve with these results shows this very satisfactorily. This gives further proof that the reaction is not bacterial.

VI. LIPASE IN SEED OILS, BUTTER, CHEESE AND CONDENSED MILK

When disagreeable flavors develop in fats and oils and in products containing them, it is of considerable commercial importance since it may involve large losses. It is also of as great theoretical interest to learn the cause of such changes. Much work has been done on the subject yet there is but little agreement of opinion chiefly for two reasons: First, investigators are prone to draw conclusions from evidence which is at once insufficient and open to objection, and secondly, it has been difficult for one investigator to know just what another has meant when he describes these peculiar flavors. For instance, investigators formerly described as rancid all unusual flavors developing in fatty and oily products in storage. More recently particularly in dairy products "rancidity" has been used to describe only that odor resembling butyric acid, and the term "tallowiness" has come into use (59). Now no doubt many flavors which are really different are being described as tallowy. Therefore, it is not possible to judge as to just what condition is meant when an author describes his product as rancid or tallowy. In this discussion rancidity will refer to an actual fat hydrolysis which in dairy products results in the liberation of free butyric acid and the development of the odor characteristic of that substance.

It would not be safe to assume that all decompositions of fat substances in storage are due to the same cause, yet it is certain that if lipase is present in fat or oil that some change will take place. Breaking down of the fat in fat-bearing seeds and seed oils has been frequently attributed to lipase, often merely

by assumption but sometimes the presence of lipase has been directly proved (60).

The decomposition of fat in milk products is frequently noted. Whole milk powder develops a flavor on long storage which makes it impossible to be used as a food. Powdered skim milk does not change in this way which is proof that the change is connected with the fat. Since the flavor is never that of butyric acid but more like tallow, this condition should be called tallowiness rather than rancidity.

Both tallowiness and rancidity are found in butter. Without doubt true rancidity in this product is accompanied by the presence of butyric and related acids. It has been believed by some investigators that these acids are produced in butter by the action of bacteria on the lactose or casein. Some have concluded that certain species of bacteria either split the fat directly, or secrete lipolytic enzymes which do. Some of these assumptions have been shown to be possible (61). Air, light, and heat also bring about changes in fats and oils though the reactions taking place are much more complex than mere hydrolytic splitting (62), oxidation of the fats and later breaking down of the molecules probably are the chief factors.

On the other hand there are many things which point to the probability of the natural lipase of the milk being carried into butter and other milk products, there splitting off butyric and other acids from the fat thus leaving the product rancid. Rogers (47) offers some experimental proof of the theory. It has frequently been shown that butter made from pasteurized cream is of better quality than when prepared from raw cream. Also when butter or butter fat is strongly heated it is found that the keeping quality is enhanced and that it does not increase in acidity as does the unheated check sample. The fact that heating increases the stability of butter fat is in support of the theory of enzymes as a cause of deterioration.

Lipase of milk is quite easily destroyed by heat. Hippius (48) has shown that the enzyme is rendered inactive when heated at 64°C. (147°F.) for one hour. These results together with some

preliminary experiments in this laboratory indicate that the critical temperature for the destruction of lipase is very near to the pasteurizing temperature usually employed in factory practice. Since this is true and since in all probability natural milk lipase plays a part in the deterioration of butter, it is clear why some investigators have found pasteurized cream to yield butter of better keeping quality, and others have seen no benefit from pasteurizing. It is probable that in some methods of pasteurization the enzyme is not destroyed. That the temperature and method of pasteurization of the cream have a great deal to do with the quality of the butter is fully discussed by Hunziker (63).

The fact also that salted butter has been usually found to withstand storage better than unsalted (63) may also be used in support of the enzyme theory. According to Rogers the proportion of salt dissolved in the water of salted butter is about 18 per cent; it was shown by Terroine (64) that 3N sodium chloride (17 per cent to 18 per cent) reduced lipase activity about one-half. All lipase in butter would probably be in the water phase and would therefore be directly influenced by the salt.

Naturally any change which takes place in butter due to enzyme must be slow. All conditions are unfavorable for enzyme activity. The fat is not in emulsion form, and as has already been stated the enzyme probably acts only at the water-fat interface and since this is greatly reduced by the agglomeration of fat globules in the manufacture of butter, the attack of the lipase on the fat-substrate is correspondingly hindered. Butter is usually kept at a temperature below the optimum point for lipase, and as just pointed out the salt may hinder the activity. The amount of lipase in the milk and cream from which the butter is made may vary also. This would be reasonable to expect if the hypothesis of Grimmer (65) is correct—that the milk enzymes are excretory either being produced in excess in the body or coming from the food. With these points in mind it can be readily understood why investigators have reached such different conclusions concerning the effect of various factors on the keeping quality of butter. Some single factor unrecog-

nized and uncontrolled might be sufficient to throw the balance in favor of enzyme activity or against it.

It has also been pointed out that changes in cheese ripening may be in some measure due to natural milk lipase (66).

Rancidity in sweetened condensed milk is not infrequently encountered and the cause has not been definitely found (67). The following experiment would indicate that this condition is due to lipase of raw milk: To different portions of good sweetened condensed milk was added 1 per cent, 3 per cent and 5 per cent of raw milk. On standing three months all samples showed the typical odor and taste of rancid condensed milk, and were high in acidity, those with the higher percentages of added raw milk being the strongest. Checks with no addition and with boiled milk added remained normal to taste and odor. Bacteriological examination indicated that bacteria could not have been the cause of the rancidity. The high sugar content of the condensed milk is sufficient to prevent bacterial growth and the investigations recorded in this paper have shown that a high sugar content does not prevent lipolytic action. Further work is being carried on in this direction but the results so far indicate that sweetened condensed milk becomes rancid due to an error in manufacturing wherein a little raw milk may pass into the vacuum pan or the finished product, and that it is the lipase in this unheated milk that causes decomposition of the fat in the main body of the batch.

SUMMARY

1. All those enzymes which induce hydrolysis of fats and the simpler esters are grouped as lipolytic enzymes. Lipase is a term reserved for those lipolytic enzymes which split the natural neutral fats.

2. In the usual methods of determining lipolytic activity, a convenient ester or fat is chosen as substrate, the enzyme is allowed to act for a period of time and the extent to which the hydrolysis has taken place is measured by titrating the acidity. A few investigators have used other methods than titration for estimating the extent of hydrolysis.

3. If the substrate is not soluble in the digestion medium then it must be well emulsified.

4. If the time and temperature are such that bacteria can multiply, it is necessary to have present an antiseptic. The ideal antiseptic would be one which inhibits all growth of micro-organisms but does not in the least check the action of the enzyme. The disagreement of investigators over the question of lipase in such substances as milk has been chiefly due to improper control of bacterial growth.

5. Investigators have been agreed as to the presence of an enzyme in milk which splits monobutyryl—monobutyrylase. Some believe they have established proof of the presence of true lipase, others have been certain that it is not there, while in recent years the tendency seems to be to leave it an open question.

6. A method for the detection of lipase is introduced, wherein is used cream of high fat content as substrate. The digestion medium is saturated with sucrose which serves as a preservative. Titration of the medium is taken as a measure of the hydrolysis. The advantages of the method are: a natural fat is employed which is well emulsified, the medium is preserved with a substance which prevents the growth of all bacteria that might interfere but which does not check enzyme action.

7. That milk normally contains a lipase which splits butter fat is proved. A few preliminary experiments indicate its resemblance to pancreatic lipase.

8. Where rancidity of fats and oils has resulted from hydrolytic splitting there is every indication that it has been caused by lipase. Since lipase has been proved to be normally present in milk, one of the causes of rancidity of butter and cheese is very probably the carrying over of this enzyme into the manufactured product.

9. Rancidity in sweetened condensed milk has been found to result when a small amount of raw milk is added to the product. This condition is doubtless due to the action of lipase of the raw milk splitting the fat of the condensed milk, the butyric acid resulting giving the characteristic disagreeable odor.

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STUDIES IN THE CONTROL OF A MUNICIPAL MILK SUPPLY

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The achievements of the past three decades in the fields of public health demonstrate the effectiveness of organized community efforts in the attainment of a healthful environment. One of the most important problems involved in municipal health administration is that of proper supervision of the supply of milk which is universally recognized as our most essential food.

The "four essentials" for the production of a clean milk of low bacterial content are: (1) Sterilized utensils; (2) small top milking pails; (3) clean milking animals and careful milkers; (4) adequate facilities for cooling and storage.

The veterinarian is an important factor for his advice in the care as well as in the testing of a herd; and the inspector is of tremendous value in emphasizing the need of cleanliness in milking, and the careful preparation of the milk by immediate straining, cooling and storage. It is obvious, then, that our information regarding the quality of a milk supply must be derived from a study of the bacterial content, coupled with inspection of the source. This is a problem of first concern, especially in our small cities. The milk supply of our larger cities, New York, for example, has, by careful supervision, improved so tremendously during the past few years that it is now usually found to be of a higher grade than that of our smaller cities. It has been demonstrated that milk of low bacterial count if properly cooled and placed in refrigerator cars may be delivered in good condition even after a haul across several states. Small cities, however, are not dependent upon milk from a distance but secure their product a few hours after milking, from close adjoining farms. Cooling and storage under such conditions will be neglected unless properly supervised.

Careful studies reported by the Virginia Agricultural Experiment Station (1911) (3), Brew (4) and North (2) have demonstrated that the standard score card is of practically no value in grading the quality of any individual milk because high grade milk may be produced from low-scoring dairies, the converse being equally true. The above observation has been further confirmed in a recent detailed study, over a period of thirteen months, of dairies supplying milk to a southern city of 37,500 population. It was found, for example, that a dairy averaging a score of 77 for the period of a year, produced milk showing bacterial counts which averaged 3,855,000 per cubic centimeter from analyses made almost weekly, ranging from individual results of 2000 to 72,000,000, whereas a dairy scoring 50½ gave one of the lowest results for the study, an average bacteria result for the same period of 250,000 per cubic centimeter, the minimum count being 1000, and the maximum 3,000,000. In the latter dairy personal interest and careful methods, factors essential to clean milk production, were highly developed, in spite of the absence of a new barn and modern equipment which the former dairy possessed. Scored on North's Score Card (2) which gives 90 per cent of the weight to the primary methods and equipment of milking, cooling and sterilizing, these dairy scores would have been adjusted to correspond more closely with bacterial results, the low-scoring dairy being raised from 50½ to 75.8.

In this study of municipal milk supply, eighteen dairies were under observation, of which number twelve were considered for the entire period from July 15, 1919, to July 31, 1920. Samples of the milk of the previous evening (stored according to a variety of conditions, sometimes iced, sometimes kept in spring water only, etc.) and of the morning's milk, which arrived within four or five hours after milking, were collected nearly every week. The samples were plated on agar according to the technique of "Standard Methods of the American Public Health Association" (1). The cleanliness of the milk was determined by the stain produced after filtering a half pint of milk through absorbent cotton discs, a perfectly clean milk scoring 100 per cent. Dairies were scored on the United States Department of Agriculture

Score Card by the city health officer, Dr. C. E. Smith, and the author.

Our studies of milk supply and dairying have led us to conclude with North that "There is no monopoly by model dairies of the sanitary methods vital to clean milk production, and consequently clean milk production is a possibility for even the poorest dairy farmer." One of the dairymen who produced the cleanest milk in this group reported kept a large herd of healthy cows, and with the assistance of his family was able to produce a clean milk with low bacterial content. He took personal interest in this milk production. On the other hand some dealers who owned dairies scoring among the highest found it necessary to employ several men, who of course did not take a personal interest in results; consequently it was impossible to produce a low count milk consistently. The "personal factor" in milk production is exceedingly important, for the care used by the milker in the dairy is one of the most important factors in the production of milk of low bacterial content. This is a factor which the present type of score card does not emphasize.

In order to obtain a low bacterial count, the dairyman must exercise care in certain essentials which have been previously suggested. The milk pails must be sterile, i.e., free from bacteria and dirt; the udders and flanks of the cow should be cleaned, preferably with a damp cloth, the air in the milking stable should be as free as possible from dust—feeding, sweeping, and passage of cows or other animals through the stable, or any other factor which would increase the amount of dust should not occur previous to or during milking; and the milk should be immediately cooled to 50°F. or lower, and kept cool.

While there may be no direct relation between individual dairy scores and bacterial counts, if we group the dairies equally according to scores we find that low-scoring dairies generally produce high count milk, as is indicated in the following table.

A similar grouping of the results of Joel (not published) (5) from a study of 125 dairies in Connecticut further confirms the above results as is shown by the following figures of average bacteria per cubic centimeter; 25,060; 27,600; 40,169; 58,950.

In other words, there is a fairly definite relation between equipment and methods and bacterial count. One seldom finds a dairyman who has dirty, unsanitary surroundings who is not careless in his methods. On the other hand, a dairyman who has a clean, modern barn and equipment is generally careful in his method.

It is not sufficient, however, for satisfactory milk supervision to employ dairy scores alone, but it is absolutely essential to carry on both inspection of dairies and laboratory examinations. Both of these are exceedingly important. Of the two, the latter is believed by the author to be of greater value for it is evident that the laboratory can test the milk of ten farms while the inspector is inspecting one.

TABLE 1

Bacterial counts and dairy scores of dairies under supervision, July 15, 1919, to July 31, 1920

NUMBER OF DAIRIES INSPECTED	DAIRY SCORES	AVERAGE BACTERIAL COUNTS	MAXIMUM DAIRY BACTERIAL COUNTS	MINIMUM DAIRY BACTERIAL COUNTS
3	77 and over	2, 122, 000	3, 855, 000	1, 015, 000
3	62-70	6, 316, 000	12, 000, 000	2, 104, 000
3	61-61.5	14, 750, 000	21, 126, 000	4, 424, 000
3	46.1-60	17, 795, 000	32, 253, 000	611, 000

The bacteriological examination of milk is the most reliable index that we have of its quality, and with this aid we can generally show the dairyman the source of his trouble. Examples of this fact are common. One instance will be cited. In March, 1920, *B. prodigiosus* was found in the agar plates, and consistently thereafter through the month of June, from milk produced by a dairy excellently equipped and supervised. On June 24 the dairy was very carefully inspected. Previous suggestions for cleaning equipment had failed to eliminate this organism. At this time it was found that a milking machine had been installed on the first of March. Special recommendations for sterilization of the machine were made and the organism was not found again. Considering the fact that the milking machine was introduced previous to the appearance of *B. prodigiosus*, and that special

attention to other pieces of equipment failed to eliminate the organism, whereas particular care in the sterilization of the machine was followed by complete disappearance of the organism, it is thought that the milking machine may have been the source of the trouble. Special care in sterilization of the machine also lowered the bacterial content of the milk. In connection with the use of milking machines the recent work of Bright at the New York Agricultural Experiment Station (11) should be mentioned. He concludes that "while machines are quite complicated, yet clean milk can be secured with them if proper precautions are taken. These precautions are such that they come within the limits of the ability of every dairyman. The all-important principles which must be kept in mind is strict attention to detail."

If a laboratory examination consistently reveals high bacterial counts, or some other factor possible of correction, an inspection of the dairy and surroundings will generally suggest the source of trouble. "The laboratory test should come first and make the diagnosis; the dairy inspector should come second and apply the remedy." Problems connected with the production and handling or distributing of milk can be controlled by education better than through legislation. Rules and regulations are essential, but the gradual education by the inspector and health officer count tremendously. The progressive dairyman also comes to regard seriously his laboratory report, and to detect the cause of a high bacterial count or dirty disc.

In a study of the relationships of groups of bacterial counts to the cleanliness of the samples, the average bacterial count was found to vary inversely as the per cent of cleanliness; and in general in our results, samples showing a sediment test of from 80 to 89 per cent clean gave a bacterial count at least from three to four times higher than samples 90 per cent clean or better. Samples having a lower sediment test showed considerably higher bacterial counts. It should be stated in this connection that this *general* principle is of only limited application because it is noted that *individual* figures for cleanliness and bacterial count do not always show such a relationship, and a milk free from

sediment may give a high bacterial count and vice versa. Campbell, in 1916, in his study of various forms of sediment tests and their relation to bacterial count concluded that "The quantity of sediment or visible dirt present on the disc is not a criterion as to the kind or number of bacteria contained in the milk." This was a study of a small number of samples which were considered individually. Whereas the results obtained in the present study confirm this careful research when *individual* samples are studied, the additional fact of a general relationship of these two factors seems worthy of note.

One of the problems in the production of milk on the farm is that of proper cooling and storage. Pease (1916) (8) in a report on a study of the relation of high temperature of milk on receipt at the creamery to the bacterial content obtained concluded that the temperature of the milk as it was received at the creamery was not a safe indication of the bacterial count of the product contained therein. In studies supervised by Conn (9) on New York City milk it was found that milk properly iced showed no appreciable increase in bacterial counts over periods of even 48 hours. Reed and Reynolds (12), in a study of milk of various qualities with agar and gelatin media, found that many organisms are able to increase in numbers after long periods even at cold storage temperature.

Extensive studies have been made to show the effect of holding milk at different temperatures during varying lengths of time, the experiments having been carried on under ideal conditions. The results of Ayers, Cook, and Clemmer (10) indicate that even if milk when fresh shows a low bacterial count the number of bacteria will be high if it is held at a high temperature. This is of especial importance in relation to night's milk. The results of the authors just mentioned further indicate that the effect of temperature on the growth of bacteria in milk during storage and transportation is important. If a low count milk is desired it must be cooled and held at 50 degrees or lower on the farm, unless it is delivered immediately after each milking.

Results of a study of temperature in relation to bacterial counts are tabulated in table 2.

From the 780 samples of milk studied above, it will be noted that the percentage of samples showing bacterial counts over 500,000 increase and those under 100,000 and 25,000, respectively, decrease in a fairly regular manner as the recorded temperatures of the milk increase. These results were obtained from both morning and evening samples as well as a few mixed samples of milk studied. By classifying the morning and evening groups separately, similar results are obtained, even more strikingly for the evening's milk which was stored twelve hours longer than the morning's milk.

TABLE 2

Percentage classification of individual counts according to temperature of milk at delivery, in groups over 500,000, 100,000 and under, and 25,000 and under: July 15, 1919 to July 31, 1920

TEMPERATURE	NUMBER OF SAMPLES	PERCENTAGE OF COUNTS		
		Over 500,000	100,000 or under	25,000 or under
°F.				
40-44	67	13	77	59
45-49	76	19	73	56
50-54	184	21	58	31
55-59	162	22	58	31
60-64	162	32	47	26
65-69	46	32	54	23
70-74	39	33	48	28
75-79	13	30	61	7
80+	31	32	32	12

There is no relationship between temperature and average bacterial counts of raw milk from different dairies on account of the various elements besides temperature which have to do with obtaining a milk of low bacterial count, for even if exceedingly good precautions be taken for cooling, if careless methods have been employed or dirty utensils used, the milk will show a high bacterial count.

Those who have studied the milk question realize that the difference between morning and night samples may be very great, especially if both are collected at a milk plant at the same time. Milk inspectors, in collecting samples for examination,

are inclined to take only one sample from a dealer's milk, regardless of whether the sample taken be one of morning's or evening's milk. One cannot estimate the quality of a milk from the results of such a collection. This factor has been noted by Dearstyne and Jones (13), who suggest the necessity of taking a composite sample of milk when grading raw milk by numerical bacterial content. It was found in the present study of several hundred counts throughout the period of thirteen months that the bacterial results of morning and evening samples varied widely in most cases. Fifty-three per cent of all the dealers gave higher bacterial results for the *evening* average.

Evening's milk is usually cooled more carefully than morning's milk. This would tend to inhibit bacterial growth. The germicidal action in milk has been studied by Hunziker (1901) (14), Stocking (1904) (15), Heineman (1908) (16), Chambers (1920) (17), and others, who have found an actual decrease in raw milk under certain conditions during the first two hours after milking. (Chambers concludes that the action is specific, depending on both the individual cow and the species of bacteria.) The counts at the end of three hours are not generally much higher than at time of milking, but will develop rapidly under favorable temperature after this time; hence the necessity for prompt cooling and proper storage. As most of the morning's milk from the dealers here reported was four or five hours old at time of delivery and the temperature usually high, there was ample opportunity for some bacterial growth. Furthermore, the milk was only delivered in the morning and there was undoubtedly a tendency to haste in milking. Unless there be supervision, the morning's milk will not be produced under as careful conditions as the evening's milk. The factor of insufficient light may also play some part in a poorly equipped dairy, for sufficient light is necessary for careful technique in milk production, and dairymen generally begin work before dawn.

Table 3 indicates relationships which may exist between morning's and evening's samples of market milk, these results being averages for thirteen months.

This table is arranged according to the evening temperature, the milk showing most careful storage appearing first. In the first place, it is to be noted that the low counts in evening's milk and most of those in morning's milk appear in the group of dealers who cooled well the evening's milk. A study of the high counts which appear in these groups will be interesting. Dealer R.'s results show frequent high counts mingled with very low counts, indicating a laxity in methods or some like factor operating on these days. Most of the high counts from dealer O appear during the first few months when the dairy received little super-

TABLE 3

Bacterial counts of evening's and morning's milk with temperatures, July 15, 1919 to July 31, 1920

DEALER	MEAN TEMPERATURE OF DAY	BACTERIA COUNT OF EVENING MILK	TEMPERATURE	BACTERIA COUNT OF MORNING MILK	TEMPERATURE
F	67	335,000	47	137,000	65
R	66	8,549,000	47	910,000	57
J	63	438,000	50	638,000	59
D	65	592,000	51	1,923,000	55
O	65	2,793,000	51	3,992,000	63
H	77	32,584,000	54	12,447,000	67
C	68	28,502,000	55	35,998,000	68
A	65	4,237,000	56	13,163,000	61
K	63	1,325,000	56	191,000	64
M	65	24,571,000	56	6,561,000	68
G	74	1,282,000	58	3,966,000	58
B	63	2,825,000	59	950,000	61

vision and when the daily temperature ranged high. After August 8, 1919, temperatures and counts were very satisfactory. The samples from dealer H were collected during the warm months, as the mean daily temperature of 77° indicates. The dairy during this time was in a filthy condition, and the only good factor operating apparently was the cooling of evening's milk. Morning's milk was not cooled satisfactorily in more than 10 per cent of the cases. While dealer C cooled the evening's milk, he made no attempt to cool the morning product. Sterilization facilities were quite inadequate and careful methods were not consistent, there being very great fluctuation in counts. On

the other hand, the low-count milk came from dairies in which conditions were favorable because of especial interest exhibited by dairyman or of this factor coupled with excellent equipment. The factor of storage is of immense importance. In studying a table like the one above, however, one must consider the various other elements which have to do with obtaining a milk of low bacterial count, for even if exceedingly good precautions be taken for cooling, if careless methods have been employed or dirty utensils used, the milk must show a high bacterial count.

Our findings indicate that excellent results may be obtained from storage in tanks of water if these tanks are iced in summer weather. Insulated tanks are superior to others. Dearstyne and Jones found a much less increase in count of the product of a dealer who used an insulated vat and renewed his water supply than in case of the dealer storing his product in tubs of water, or in the uninsulated concrete vats in which no renewal of water was made. Kelly (18) states that a 10-gallon can of warm milk precooled with water at 55°F. and set in a tank of ice water at 37°F. can usually be cooled to 50°F. in about twenty minutes. The best and quickest way to cool milk is by use of a surface cooler with the coldest available water, then setting the cans in a well-insulated tank of ice water. Extreme care must be exercised in the cleaning of such a cooling apparatus. Gamble (19) found that felt jackets or insulated cans proved to be very effective in keeping milk cold during long shipments in hot weather and in preventing freezing during cold weather.

The seasonal variations in bacterial counts in milk have been frequently noted by North and others, who have observed that winter weather produces a reduction in bacterial counts. In table 4 there is presented the average monthly results of our study from July, 1919, through July, 1920. The mean temperature for the days on which milk was sampled were kindly furnished by the United States Weather Bureau.

A study of the above results indicates a rather striking seasonal variation in bacterial counts obtained from an average of *all results* from the dealers under supervision as well as from an average of the *maximum* and *minimum* counts for each month;

for all these counts become lower during the winter months, increasing again with the approach of hot weather. The column of mean daily temperatures is interesting in this connection, because in this way the possibility of warm days in the winter season (not uncommon in this locality) is taken into consideration. Another outstanding feature shown in the table is the appearance of uniformly high bacterial counts during the first two months of this investigation, those for July and August being enormous. For the corresponding month the year follow-

TABLE 4

Monthly results: Bacteria per cubic centimeter, average, maximum and minimum; temperature of the milk and mean temperatures of the sampling days, July 15, 1919 to July 31, 1920

MONTH	NUMBER OF SAMPLES	MEAN TEMPERATURE IN DAYS	AVERAGE TEMPERATURE OF ALL SAMPLES	AVERAGE BACTERIA PER CUBIC CENTIMETER OF		
				All counts	Maximum counts	Minimum counts
July.....	38	79	60	55,464,000	160,222,000	500,000
August.....	54	80	59	69,356,000	197,600,000	184,000
September ...	48	76	55	937,000	4,690,000	11,300
October.....	49	70	57	162,000	772,600	11,300
November.....	55	55	54	140,000	832,000	3,200
December	21	61	57	458,000	1,227,000	22,000
January.....	50	54	56	258,000	1,611,000	3,200
February.....	62	44	48	180,000	552,000	3,100
March.....	45	52	54	111,000	502,700	2,400
April.....	47	59	56	109,000	934,000	36,000
May.....	14	67	57	340,000	403,750	278,000
June.....	43	74	68	704,000	1,748,000	39,000
July.....	38	82	57	799,000	2,160,000	43,000

ing, the average counts do not even approach such high averages. Inasmuch as the results in 1920 are obtained from the same dealers studied in 1919, and practically the same number of samples were tested from each of the dealers throughout the year, it is safe to conclude that season is one important factor in the low counts of the cooler months. Furthermore, as there was no supervision of these dairies for a few months prior to July, and as the counts show such a decided drop and do not again return to such high proportions, it may be safely assumed

that the reduction of these high counts is due in a measure to supervision by laboratory analyses and dairy inspection. The ordinary dairyman, whether consciously or not, will strive harder to produce a milk of low bacterial content if he knows that his milk is subject to laboratory examination.

SUMMARY AND CONCLUSION

If the public health laboratory be used as an aid in determining the quality of a milk supply, and the results carefully analyzed in connection with the inspection of the source of supply, a reliable estimate may be obtained of the quality of the product from the dairy. The dairy inspections should be used entirely as educational measures. The laboratory indicates whether or not the milk has been adulterated and is an index of methods employed, while the inspection will place the blame where it belongs. In general, the better class of dairies produce the higher quality of milk, but this rule does not hold in all cases, for we frequently find a low-grade dairy producing a clean, low-count milk. In such cases the methods employed are good and offset the lack of equipment and sanitary surroundings. The tools of the laboratory and dairy inspection are invaluable for the health officer in the safe-guarding of a milk supply, and constant supervision of this nature will insure a more satisfactory product.

In the sampling of raw milk supplies, it is essential to collect samples both from the evening and morning product for a fair estimation of any day's milk. If careful supervision be given the milk production, and factors of cleanliness, sterilization, use of small-top pails, healthy cows and milkers, cooling and storage be employed, a consistently low-count milk may be produced. All of these factors are vitally important in the production of milk which must be stored for any period. Temperature has a very decided effect upon bacterial growth, and the bacterial counts invariably increase during the summer months in the milk of those dealers who do not carry out systematic cooling. Proper cooling, however, will maintain low bacterial counts. It has been found practicable by those dairymen who are personally interested in the production of a good milk supply to cool the

morning milk to 50°F. and maintain that temperature until delivery.

Many dairymen today have studied their problem, are well versed in the matters of health and sanitation, and desire to deliver a good product. This is especially true of those who maintain large routes in the cities. Many small dairymen, however, some of whom deliver their milk at wholesale to creameries and milk plants, are either new in the business or have never given serious consideration to the milk problem. If the health officer or milk inspector is tactful in approaching this type of dairyman, he may accomplish remarkable results in the way of education and response.

The best solution we have at present of the municipal milk problem is a grading system based on dairy scores and bacterial counts, together with pasteurization of all milk except that of the very highest grade.

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A COLOR TEST FOR "REMADE MILK AND CREAM"¹

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Remade milk and cream are notable additions to our list of dairy products. The increased use of these products calls for a method of distinguishing them from natural milk and cream in order to protect the producer as well as the consumer against fraud.

A report (1) has already covered an investigation of several methods of attacking this problem. These methods were found to be of little or no value in detecting mixtures of natural and remade milk. A practical qualitative test for distinguishing natural pasteurized milk from mixtures of natural and remade milk is reported here. As little as 10 per cent of remade milk, varying somewhat with the grade of powder used, can be detected by this means. If condensed milk has been used in making the remade milk, the amount that can be detected will depend upon the degree of heat to which the condensed milk has been exposed in the process of manufacture.

According to Porcher (2) and Cazalas (3) no change has taken place in the casein of milk powder in the course of its manufacture except for a loss of sulphur. Jensen (4) states that the effect of heat on milk is to brown the casein, while Cazeneuve and Haddon (5), Leeds (6) and Hotz (7) claim that the lactose is altered. An increase in the titratable acidity of milk when exposed to a high degree of heat was noticed by Jensen (8) and Steinegger (9), while Van Dam (10) and Milroy (11) found an increase in hydrogen ion concentration. The action of caustic alkali on pure lactose was studied by Framm (12), Schade (13), Meisenheimer (14), Nef (15) and others, and its action on whole milk by Kruger (16), Gautier and Morel (17) and Grimmer (18).

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In the course of experiments made in this Laboratory on the effect of alkali on the curd of milk, it was discovered that when the washed curd of the product made from milk powder was dissolved in sodium or potassium hydroxide, a yellow color developed in a few hours. It was also observed that the washed curd of natural pasteurized milk did not give this color.

It is known that pure lactose will give a yellow color when dissolved in caustic alkali. This is known as the "Moore test" (19). Five milligrams of lactose is sufficient to produce a trace of color in 10 cc. of 5 per cent sodium hydroxide. A yellow color is also produced by dextrose, fructose and maltose, but not by sucrose. The color is attributed to the aldehydic or ketonic nature of these sugars. It is also known (20) that aldehydes combine with proteins, probably with their free amino groups (21). According to Steinegger (22) the increase in acidity of milk when formaldehyde is added is due to a combination of the CHO group with an NH_2 group, with the elimination of water. It is conceivable that in the course of the manufacture of milk powder, lactose, being an aldehyde, might under the influence of heat and desiccation combine to a slight extent with casein and albumin in the same manner as other aldehydes. Experiments made in this Laboratory showed that there was a slight decrease in per cent lactose, as obtained by copper reduction, when milk was heated in boiling water for one and one-half to two hours. A slight increase in titratable acidity was also noticed which, however, may be explained in other ways (23). The yellow color produced by the action of alkali on the curd of remade milk, is, therefore, probably due to the presence in the curd of a compound of lactose of such a nature that it is not removed by washing. This compound has been formed presumably by the action of heat and desiccation in the process of manufacture of the milk powder and may possibly consist in a linking of the lactose with the protein, the aldehydic group of the lactose losing an atom of oxygen and the amino group of the protein losing two atoms of hydrogen.

The samples used in this investigation were made from skim

milk powder and unsalted butter as well as from whole milk powder and the components were so chosen as to bring the fat and non-fatty solids within the range for natural milk. Two types of emulsors and an homogenizer were used. Spray powders from eleven different manufacturers and drum powders from four different manufacturers as well as different samples and brands from the same manufacturer were used.

A few commercial samples of remade cream and ice cream in which milk powder had been used as well as 70 samples of market milk representing different distributors in the cities of Chicago, New York, Philadelphia, Baltimore, Washington and Richmond, were tested in the manner recorded here. Those samples which, from the evidence available, were known to contain remade milk or cream, responded to the test.

Experiments were made to determine the effect of heat on natural milk and it was found that in milk heated at the temperature of 70° to 72°C. for thirty minutes, the curd, when treated with alkali according to the method recorded here, gave a slightly different shade from that of milk heated at 63° to 65°C. for thirty minutes. The former temperature, however, is considerably above the holding temperature of pasteurization. A temperature of 80°C. for one minute was found to have no appreciable effect. It may be mentioned here that the peroxidase test was used in connection with these experiments and also on the commercial samples of market milk tested. This test begins to weaken or disappear at the temperature of about 72°C°. All the samples of market milk examined gave a positive test for peroxidase. This test was made in two ways as follows: To 10 cc. of milk were added 5 drops of 1 per cent tricesol (24), 5 drops guaiac reagent (4 grams guaiac wood in 50 cc. acetone) and a drop of commercial H_2O_2 . To another 10 cc. portion of milk were added 2 drops 1 per cent benzidine in 50 per cent alcohol, 2 drops 1 per cent α -naphthol in 50 per cent alcohol and 1 drop of commercial H_2O_2 . A blue color develops in the former case and red in the latter, provided the peroxidase has not been destroyed by heat.

METHOD FOR MILK²

To 25 cc. of milk in a 250-cc. beaker was added an equal volume of distilled water and after warming to 25° or 30°C. the curd was precipitated with 3.5 or 4 cc. of 10 per cent acetic acid. Distilled water to the amount of 200 cc. was then added and after standing for some time to settle, as much as possible of the supernatant liquid was decanted through a 166-mesh silk bolting cloth. The curd left on the cloth was washed back into the beaker. The beaker was again filled with water and the curd allowed to settle and decantation made as before. This was repeated three or four times. The curd was then transferred to a 15-cm. rapid double filter and washed at least three times, filling the funnel nearly full each time. The curd was broken up with a glass rod to facilitate washing. The filter was then removed from the funnel and after squeezing out the excess of water with the hand, the curd was placed in vials of clear glass 17 by 100 mm. and 10 cc. of 5 per cent sodium hydroxide added, the curd being broken up and mixed with the liquid with the aid of a glass rod. The yellow color began to develop in about two hours, the vials, however, were left over night and the final observation made the next day.

The control or standard for comparison was a sample of natural milk pasteurized at the temperature of 63° to 65°C. for thirty minutes. Several 25 cc. portions of this control were treated at the same time in exactly the same manner as the unknown and placed in vials of the same diameter.

METHOD FOR CREAM

To 15 cc. of cream was added an equal volume of water and after warming to 30° to 35°C., the curd was precipitated with about 2 cc. of 10 per cent acetic acid, filtered and washed. The greater part of the fat was then removed by washing first with 25 to 40 cc. of 95 per cent alcohol, then with 50 to 75 cc. of pure acetone using small quantities at a time, the curd particles being

² If the milk has been homogenized, the fat should be removed as in the method for cream.

broken up with a glass rod after each addition of the solvent. The curd was then washed thoroughly with water to remove the acetone and after draining was placed in glass vials of the same size as those used for milk, and 10 cc. of 5 per cent sodium hydroxide were added.

The standard used for comparison was a sample of natural pasteurized cream of about the same fat content. No attempt was made to detect mixtures of natural cream and remade cream.

SUMMARY

A practical qualitative method has been given for detecting remade milk and cream, based upon the color produced when the curd is dissolved in sodium hydroxide.

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METHODS OF CARING FOR MILKING MACHINE TUBES

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At a Milker Conference held at the New York Station on May 27, 1921, one of the most important points discussed was the relative value of the hot water method of sterilizing the teat cups and tubes, and the chemical method of sterilization by means of harmless chemicals such as brine and hypochlorites. Because the Station has frequently been asked to discuss its experiences in this matter, the following statement has been prepared.

The most natural thought in connection with the cleaning of any dairy utensil is that of scrubbing it, following this procedure with scalding water or flowing steam, and completing the cleaning process by drying. It is, therefore, not surprising that practically all of the early investigators of sanitary methods of cleaning milking machines (Harrison (6), Stocking (12), Stocking and Mason (13), Edwards (3), Meek (9), Haecker and Little (7), Harding, Wilson and Smith (5), Hoffman-Bang (8), Williams, Golding and Mackintosh (14), Burri and Hohl (2), and others) tried this method for milker tubes. Where the heat used was less than the amount necessary to kill microorganisms present, or where other sources of contamination were left uncontrolled, the results were very unsatisfactory. The bacterial counts under these conditions were usually high. Where sufficient heat was used to sterilize or to practically sterilize the various parts of the machine, the results reported are better and in several cases excellent. Thus for example, Edwards (3) reports counts as low as 1407 and 1776 per cubic centimeter when the tubes were thoroughly cleaned, boiled and steamed.

Practically all of the earlier investigators who have used heat-sterilization mention the fact that the rubber parts are so rapidly destroyed as to make this method of sterilizing milker

teat cups and tubes by boiling water or by steam impracticable under ordinary conditions.

With the development of the rubber industry and the development of heat resistant rubbers for use in surgical gloves, automobile tubes and other things, this situation has changed and there are today several rubber manufacturers ready to supply rubber parts for milkers that are designed to withstand boiling water or steam and that are at the same time sufficiently elastic to serve the purposes of mechanical milkers. This is particularly true of cloth-wrapped rubber tubing. Rubber parts such as are necessarily made in moulds are apparently not yet made of a truly heat resistant rubber; so that those milkers using moulded rubber inflations, or moulded rubber mouth pieces, are placed at a disadvantage in the use of the heat-sterilization method. Yet it may justly be held that moulded rubber parts for teat cups should not be discriminated against as it is usually so difficult to remove the straight rubber tubing inflations from the teat cups that companies using this type of inflations do not ordinarily advocate the complete removal of these inflations except for renewal, and provide for cleaning them without removal. Moreover, all persons familiar with the details of the matter seem to agree that even those machines which are normally equipped with heat resistant rubber inflations and tubes occasionally receive a lot of rubber parts from the manufacturers of these goods that are of very poor quality. This condition of affairs is apparently due to the difficulty that manufacturers of rubber goods have in supplying material of an absolutely standard and unvarying quality.

In view of the fact that several of the standard makes of milking machines in general use on dairy farms are not equipped with heat resistant rubber parts, it is unfortunate that sweeping statements have recently been published as to the general applicability of the heat-sterilization method for caring for the teat cups and tubes of milking machines, some investigators even asserting that this method of sterilization is the only method that can be used with good results.

Because of the difficulty in getting rubber goods of the right quality for use where heat-sterilization was to be used, investigators have from the first tried to secure satisfactory methods of sterilizing the rubber parts by means of chemicals of various kinds. Thus among the early workers in this field, Erf (4) tried boracic acid, a solution of lime, and formaldehyde. Stocking (12) and Stocking and Mason (13) tried brine, borax, lime water, formaldehyde, and soap powders, while Harding, Wilson and Smith (5) used brine. From this early work it became evident that brine was the most satisfactory solution that could be used for keeping the tubes sweet and clean, but its use was accompanied by the difficulty that some metals were corroded by it. Also, it was a preservative rather than a sterilizing agent. It was not until Ruehle, Breed and Smith (11) showed that the brine organisms were not capable of growth in milk and that milk organisms were not capable of growing in brine, and Wing (15) showed that brine could be readily and efficiently sterilized by the use of hypochlorites that the value of the brine-hypochlorite combination became really evident. Meanwhile an active advertising campaign was started by various firms selling hypochlorite solutions urging that milking machine tubes be disinfected by these solutions used alone. The net result of the latter campaign has been one of great disappointment. While milking machine tubes can be effectively sterilized by the use of hypochlorite solutions alone where these are used in sufficient strength and the strength is renewed with sufficient frequency, (Ruehle, Breed and Smith (11)), very few users of milking machines have appreciated the limitations of this type of sterilizing agent well enough to succeed continuously in producing a low count milk. In those cases where dairymen have neglected these solutions, it has not been uncommon, especially in hot weather, to find a man keeping his milker tubes in a stinking solution entirely free from any sterilizing agent and full of enormous numbers of organisms. Probably no one thing has so delayed the day when milking machine users will get satisfaction from their purchases and continuously produce a milk of good sanitary quality, as has this campaign of commer-

cial firms for the use of hypochlorite solutions. Hypochlorite solutions are highly effective for use with a preservative solution like brine; but when used alone, they require more attention than the average dairyman can or will give them.

However, the limitations of the brine-hypochlorite solution are such that no recommendation should be given for its universal use. The majority of the teat cups of milking machines are now made of metal parts that are not corroded by this combination; but in certain types of machines, because of mechanical limitations, this has not yet been accomplished with entire satisfaction.

A third method of caring for milker tubes has also been used quite commonly and with fair success in New York State where really cold springs are quite common. This method consists of allowing cold water to circulate through and over the milker tubes and cups between milkings. It depends for its success upon general cleanliness and the retardation of bacterial growth through the effect of cold and is successful only when low temperatures (preferably less than 50°F.) are maintained. Because this method of caring for the teat cups and tubes can be used without injury to rubber parts or to the metals ordinarily used, it has been tried in some cases where conditions did not justify its use. It is not a positive method of sterilizing in that no bacteria are killed, and results can never be made as perfect as in those cases where proper heat methods or chemical methods are used. Nevertheless, there are some men who are getting good results with it in New York State under farm conditions.

With all of the procedures that have been suggested for caring for the tubes there has been a common tendency for dairy-men to rely either on heat or cold or the action of the chemical solution to destroy the bacteria, to the neglect of actual cleanliness. Milk has been allowed to dry onto the interior of the tubes, the teat cups claws have been allowed to become badly clogged with milky accumulations, check valves on the pail cover have been left uncleaned, tubes have been thrown carelessly into the solutions so that entrapped air prevented the action of the sterilizing solution, and many other details have

been neglected, with the thought that the heat, cold or chemicals took care of everything.

It was shown early in milking machine investigations that the tubes and cups could be kept in clean and sanitary condition without taking them apart daily if washed thoroughly by drawing an abundance of cold and hot water containing alkali cleaning powders through these parts *immediately* after each milking (Harding, Wilson and Smith (5)). The publication of this statement has been used as an excuse by milker salesmen and dairymen for saying that all the cleaning necessary was to draw a pail of cold water through the tubes whenever convenient after milking. In many cases this is all the cleaning milking machines have received for months at a time. This has been the case in spite of the fact that even where the tubes are cleaned thoroughly after each milking, there is always some blackening of metal parts where these are in contact with the rubber so that to keep them really clean and shining, they must be taken apart and each part individually polished at least as often as once a week. The failure of milker companies to teach the purchasers of their machines good cleaning methods and the failure of dairymen to realize the necessity for making satisfactory provision for proper conveniences for cleaning their machines had led to the production of large quantities of milk containing excessive numbers of bacteria. Largely for this reason, with the return of more abundant farm labor, the hand milker has again become a severe competitor of mechanical milkers. It is therefore not surprising to find the various milker companies coöperating in pushing a vigorous campaign for the better care of their machines by dairymen.

Recently some public health authorities, because of a very natural and well grounded prejudice against the use of chemical sterilization of dairy utensils, have threatened to forbid the use of chemical sterilization for milking machines. Fortunately, so far as known to the writer, this policy has never been put into force, and it is to be hoped that it will not be. Health authorities have every reason to be active in compelling dairymen to produce a clean milk containing few bacteria or discard their

machines; but any attempt to enforce such a regulation as indicated would take us back to the days when dairymen were instructed by control officials that clean and sanitary milk could only be secured in whitewashed barns, with a specified number of windows and so on. Under present conditions, if health authorities or investigators dictate what method of cleaning milkers shall be used, or state that only one method is successful, because of mechanical limitations in the construction of milking machines, it gives the support of public agencies to one group of milker manufacturers as opposed to a second group. If there were any danger involved which affected the public health such a course might well be justified, even though one group of commercial interests were favored; but there is no evidence at present available that indicates the presence of such a necessity.

Some investigators have not realized the difference between the use of the ordinary chlorine solutions and the use of the brine-hypochlorite solution, or have reported that the latter method was not successful because they knew of instances where dairymen had reported that they were using it, yet the results secured were unsatisfactory. In the latter cases (as in the instances reported by Bright (1)) an investigation would undoubtedly show that while the dairyman may honestly think he is following the directions, he is failing to observe some essential step in the procedure.

Inasmuch as it has been amply shown that the chief source of the bacterial contamination of milk is from the dairy utensils with which it comes in immediate contact, (Prucha, Weeter and Chambers (10)) and as literally tens of thousands of dairymen in the United States, are using milking machines, the matter of a campaign for better care of these machines is highly important to all of the parties concerned. The public is interested through its agents, the public health authorities and experiment stations and colleges. The dairymen themselves are interested because any tendency to lower the quality of dairy products injures their business. The milker companies likewise face the necessity of showing that milking machines are capable of giving satisfac-

tion in the hands of all, or practically all, purchasers of their machines, or their business will disappear. Now that proved and tried methods of cleaning milkers are known, it would appear that the time is ripe for more vigorous campaign measures by all of these forces to improve a situation that is not what it should be. The milker companies, as already indicated, are individually or collectively organizing campaigns along lines that should command the support of everyone. Some states, such as New York, are carrying information directly to dairymen through extension activities. Some city milk inspectors and inspectors of dairy companies are securing correct information regarding these things and carrying it to the dairymen with whom they come in contact, and some of the dairymen's organizations are taking a real interest in encouraging their members to produce better quality products. Proper coördination of these activities would hasten the day when the users of milkers will clean and sterilize them properly. In New York State gratifying results are already evident from coördinated efforts along these lines so that we already have milk plants where educational measures have so reduced the trouble from dirty milking machines that many users of machines are continuously securing premiums at Grade A milk stations for the production of milk with a bacterial count less than 10,000 per cubic centimeter.

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OUTLINE FOR A STUDY OF THE COST OF MILK PRODUCTION

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More studies of the cost of production in the dairy business have probably been made, than in any other phase of agriculture. Yet the dairy farm is one of the most complex of agricultural businesses. The reason for the great number of investigations into the dairy business is mostly that, many farmers felt¹ they were losing money on their dairies, and the dairymen were among the most vociferous of all farmers in demanding higher prices, or some sort of a change in the conditions of the business so that their dairies might become paying propositions.

The writer has examined critically, the methods and the conclusions of over twenty investigations into the cost of milk production. These reports or bulletins cover practically all the important milk producing sections in the country. It was found that the methods of collecting the data, the methods of computation, and the systems of conducting the studies were almost as many as the reports. On a few of the more important and outstanding factors in the production of milk, the different bulletins were pretty well agreed, but as regarded the greater number of factors, very few of these bulletins could be compared directly, one with another. A careful study of a whole bulletin, and of the methods of collection and tabulation of data was always necessary, in order to understand fully the significance of its conclusions on any particular factor of cost.

The writer has endeavored to pick out, from among these studies, the methods of collection of data and of computation of each factor which seemed most reasonable, and which seemed to be based on the best business practice for the farmer.

In making a study of the cost of milk production, the herd will generally serve as the basis for study and comparison. The cost

¹ Particularly from 1916 to 1920.

will generally differ more between cows in the same herd, than between dairies, but one of the principal aims of such an investigation is generally to compare and to choose from among the different systems of management, and this can be done only when the dairy rather than the cow serves as the basis for comparison. Hence, it is the comparison of dairies rather than of cows that is considered in the following pages.

COLLECTION OF DATA .

There are several possible methods of collecting data for an investigation of the cost of milk production, but the principal ones may be grouped under two headings; the Survey Method, and the Cost Accounting Method.

The survey method

The survey method is the one which will generally be used when the nature of the study does not allow enough time, or when the financial provisions do not permit of a sufficient expenditure for the use of more careful and accurate methods of collection.

Under the survey method, a comprehensive questionnaire is first drawn up, and a corps of men who are thoroughly acquainted with the dairy business, are employed to visit the farms. These men, by conversing with and questioning the farmers get the information necessary to fill out the blanks.

A great deal of care must be put on the questionnaire before any data is collected. Some of the principal points to be borne in mind, are the following:

1. The questions must be so clear as to be proof against misunderstanding or confusion on the part of the dairymen.
2. They must be so formulated that there will be no possibility of their antagonizing the farmer.
3. The questions must be in the farmers' vernacular, and free from scientific abstractions.
4. The questionnaire should cover every factor of the business that could possibly influence the facts which are being sought.

5. At the same time, useless questions must strictly be kept out, and the questionnaire should not be so long as to tire the farmer before he has discussed all the important factors.

6. Where it is possible, check questions should be incorporated in the questionnaire, in order to verify important points.

The man who is collecting the data, must always be able to supplement the farmers' answers with observation. He must be able to use tact and diplomacy in questioning further, on points where the farmers' answers appear to depart from the facts.

In the survey method, the data from individual dairies may have a considerable factor of error, because of poor accounts, and the general proximation of answers. However, if a sufficiently large number of dairies be visited, the errors from some may be considered as offset by errors in the opposite direction from others. This is only a rough and approximate sort of a tendency, but it may be depended on to secure reasonable accuracy in a study of a large number of dairies.

The cost accounting method

This is essentially the method used in cow testing associations. It has the advantage of greater accuracy than the survey method but the disadvantage of greater expense, and usually of a decided limitation on the number of dairies from which data can be collected.

Under this method, it is necessary to keep accurate accounts for each dairy, to weigh milk and feed, watch closely the time spent on the dairy by the farmer and his hired men, and in short, to check up all factors of expense and income. It has seldom been found possible to depend on the farmers to do this work for themselves with any degree of accuracy. The best method, and the one generally used to reduce expense to the bureau making the study, and to insure accuracy and uniformity of data, is to organize so-called Cost of Production or Cow Testing Associations, in which the farmers agree to pay the salary of an experienced man who will spend at least one day a month at each farm. There it is his duty to check up the farmer's accounts, weigh milk and feed, test milk, etc. It is not necessary to go

into the details of organizations here. They are widely known, and though they vary in details, their general scheme is pretty uniform, and their principal aim is to collect more accurate data regarding the operation of dairies than the dairymen could or would be likely to do for themselves.

Among the greatest advantages of these associations are that they permit an accurate inventory to be taken at the beginning and end of the year, and facilitate the keeping of a fairly complete set of accounts. For accuracy these accounts can best be kept by the central bureau or experiment station, the farmers mailing in a complete report of the farms' or dairies' operations each day. This is entered under the proper accounts by the central bureau. The books are then balanced at the end of the year, and the complete and fairly accurate data on each factor then becomes available.

As was mentioned above, the principal disadvantage of this system, aside from the expense, is in the limitation of the number, and also of the type of dairies from which data can be obtained. In the first place; one route man cannot attend to more than twenty-five dairies. And for a comprehensive study, data from two or three hundred should be available, or in other words, from eight or more associations.

Only the most efficient and profitable dairies of a given neighborhood are likely to enter such associations, indeed, only they will be likely to pay the necessary expenses of the cow testing associations. This will leave a large proportion of the dairies out of the study, and the most unfortunate thing is that, these dairies will comprise more or less definite classes which should be compared to the more efficient ones in the association.

The cost association method is by far the more accurate, but its conclusions, based on the association records, must always be definitely limited to the types of dairies actually found in the associations, and not stated as general or sweeping conclusions.

CLASSIFICATION AND COMPARISON OF CLASSES

After the data are collected, it becomes necessary to divide them into groups or classes in two directions; first, in such a way

that all the data in any one class will be homogenous, that is it must be from dairies of similar types, operating under similar conditions, in a restricted area, and producing like products. Secondly, the data must be classified in such a way as to facilitate the study of the particular items under discussion.

HOMOGENEITY OF DATA

The business of dairying is widely affected by the size of dairy, by the type of farming common in a given neighborhood, by the proximity of markets, the disposition of the products, the physical features of the locality, and by a number of other factors. Hence, if such a study is to be of any real significance, it must be confined to a sufficiently small area so that the dairies will all be comparable. It must further, contain a clear classification of dairies according to the accompanying system of farm management, size of dairy, disposition of product, efficiency of management, etc. The extent to which this classification should be carried will depend on the aim of the study, and the factors to which particular attention is being paid. To compare New England dairies directly with dairies located in Wisconsin, to include in the same table large and small dairies, or dairies selling "A" grade milk with those making butter, is to invite error. And to draw any but the most general conclusions from such data is fallacious and may be even harmful.

FACILITATION OF THE STUDY

The final comparison in a cost of production study will usually be based on the averages of the different classes of dairies from which have been drawn the data at hand. Considerable care must be taken in forming these classes in order that the averages may be comparable.

There should be a sufficient number of dairies in each class so that errors in collection and tabulation may have a chance to counterbalance, and in order that exceptional cases will not unduly influence the averages of the classes of the conclusions of the study. A class which contains fewer than ten or twelve

dairies will not fulfill this condition, and no very sweeping statement should be based on such a classification.

The ideal situation would, of course, be to so classify the dairies that only two factors would vary from class to class—one being the variable and the other the factor in which it is hoped to trace the effect of changes of magnitude in the variable. But it would be both difficult and expensive to collect data from a sufficient number of dairies so that each class or group contains dairies, which differ in two factors only, and agree in each of the twenty or more other factors of production. Therefore, it may sometimes be necessary to include in a class, dairies which agree in the most important factors but differ in a few minor ones besides that particular factor which is being treated as a variable. In this case the dairies should be so arranged that variations resulting from differences in minor factors will counterbalance, and so will not affect the results.

The comparisons between the different classes will usually show themselves as correlations. The correlations may be discovered by inspection, by the value of a factor rising or falling consistently from class to class as the independent variable is increased or diminished. The simplest correlation would be between two factors, the one being treated as the independent variable, which is the basis of classification and hence increases at a given rate from class to class. The other, or dependent variable, will show correlation if it increases or decreases at some ratio corresponding to that of the former. For instance, in studying the relationship between the production per cow and cost per quart; production per cow may be used as the independent, and the cost per quart as the dependent variable. The dairies might be classified as follows:

- Class I, Dairies producing 2000 to 2999 pounds per cow
- Class II, Dairies producing 3000 to 3999 pounds per cow
- Class III, Dairies producing 4000 to 4999 pounds per cow
- Class IV, Dairies producing 5000 to 5999 pounds per cow
- Class V, Dairies producing 6000 to 6999 pounds per cow

Now the dairies are grouped under these classes with regard to production only, but if a correlation exists the average cost

will be found to rise from class to class as production is increased. Or if an inverse correlation exists, the cost will fall from class I to class II, from class II to class III, etc. Correlation coefficients may be derived for the use of the readers better acquainted with statistical methods.

It is assumed that before the above classification is made, the data included in it is homogeneous and agrees with the rules for homogeneity suggested above. If not, a further division to comply with these rules will be necessary. If the cost per quart varied according to size of dairy, each of the above classes should be subdivided somewhat as follows:

Class I, Dairies producing 2000 to 2999 pounds per cow	{ A. Dairies of fewer than 20 cows B. Dairies of 20 to 39 cows C. Dairies of 40 to 59 cows.
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The extent to which this sub-classification should be carried will depend, as was said above, on the nature of the particular study which is being made. In practice it is seldom possible to eliminate all but the two factors whose correlation is being considered, and it is usually necessary to permit variation in more than one factor at a time. It is best to construct the tables so that all the factors in which correlated variations could occur, will be shown in each table. Thus, it becomes possible, by cross comparison to make allowance for intercorrelation among the different factors in each class.

GENERAL FACTORS INFLUENCING THE COST OF MILK PRODUCTION

Production per cow

It is well known that the cost of producing milk, per quart or per hundred pounds, will vary inversely with the production per cow. In most investigations into the cost of producing milk, the production per cow has been made a basis for classification. In any case, it must plainly be taken into consideration, and where a comparison is made between two dairies or groups of dairies, the statistics presented should show clearly the production per cow in each dairy or class.

Size of herds

The size of herds, like production per cow, has been shown to have a strong influence on the cost of producing milk. In any study of milk production, the size of herds must be taken into consideration, and should usually be made a basis of classification. In any event, data from dairies of widely differing sizes should not be massed, and the effects of variation in size within and between different groups should carefully be watched for. The maximum and minimum sizes should be noted in summaries as well as the average or prevailing size.

Disposition of product

The disposition of product may prove a very potent factor, and should always be given some thought and mentioned in the explanatory paragraphs.

Where a study includes dairies whose products are disposed of in different ways, as for instance; market milk, butter, cheese, "A" grade milk, and "Nursery" milk, the differences in methods of dairying arising from differences in products should never be overlooked. And where any considerable difference in cost or method of management is found as such a result, the dairies should be classified according to their products.

Value per cow

The man who collects the data should have had enough experience in dairying, and should be sufficiently well acquainted with the value of cattle in the neighborhood where he is working, to be able with the aid of the farmer to place a fair valuation on the cows in each dairy.

Cows recently purchased should, of course, be valued at the price paid. Others should be valued at what they would sell for as dairy cows in that neighborhood.

In placing a valuation on the cows, their productive capacity and the likelihood of their being profitable milk producers, should be the paramount consideration. It has even been suggested^a

^aBureau of Statistics, U. S. Dept. of Agriculture, Bulletin 88.

that the profit from the cows' product be capitalized as a method of evaluation. Under this method, a cow that caused a loss to her owner might be considered a liability instead of an asset. While it may be said that there is a sort of justice in this suggestion it must be remembered that even though a cow may have no value as a milk producer, she will still sell for something for beef. And there is capital tied up in her whether it be a profitable investment or not.

Pure bred cows should be valued, for the purpose of the ordinary cost of production study, at the same rate as grade cows of similar conformation and productive capacity. If a cow has a value, *inherent in the fact of her registry, above that properly pertaining to her as a milk producer*, this may properly be left out of account in such a study. In the first place, such a value does not bear any relationship to her efficiency as a milk producer, and in the second place, it may be assumed that she will impart this value to her offspring. The higher value of her calves will offset the interest on the higher value of the cow.

OVERHEAD EXPENSES

Overhead expenses are those which continue regardless of the amount of milk produced in a dairy, or of the intensity of the business. A fundamental distinction must be drawn between these expenses and those which may ordinarily be expected to increase with the amount of milk produced, and with the extent to which the dairy is forced toward its maximum limit of production.

This list will include the following factors of cost, which are of sufficient importance to merit their being discussed at some length:

1. Interest on value of cow.
2. Depreciation on cow.
3. Maintenance of buildings.
4. Maintenance of equipment.
5. Bull service.
6. Cost of bedding.
7. Miscellaneous overhead.

Interest on value of cow

The farmer may properly be allowed interest on the capital he has tied up in his dairy, whether in cows or equipment. If he had no capital of his own, he could not conduct nor begin his business until he had obtained it, either from his own savings or in the form of a loan from someone else. And a man who has capital cannot be expected to put it in a dairy unless it can be expected to return him as high a rate of interest there as if he bought sound securities, or put it in some other business. The rate of interest allowed the farmer on his investment in his farm or dairy should be the same as that at which he could borrow it, which would usually be at the rate of short time loans in the neighborhood.

Interest on the investment in cows constitutes more nearly an overhead charge than a secondary or current one, for the investment in cows is subject to no variation whether the cows be forced to their maximum production or not. Interest on the feed kept on hand, is however, more nearly a variable charge, for the amount of feed kept may be expected to vary with the amount of milk produced and the extent to which the dairy is forced.

Depreciation on cows

This is a much argued point, and several methods have been used in attempts to determine it. There have been two principal methods:

1. *Depreciation on the individual cow.* This is a theoretical method, whereby the difference between the value of the cow when she enters the herd, and that at death or sale is distributed over her period of usefulness—real or estimated. The weakness of this method, as it has been used, lies in the fact that the cow's period of usefulness was usually estimated rather than real. There has been a wide diversity of opinion as to what the usual period is. The usual estimates vary from five to eight years. Also there is a tendency to try to predict what a cow will probably be worth at a given age, rather than to base the final price on that of the cows actually disposed of.

Depreciation will vary with the original value of the cow, and inversely with the length of her life, but it must be remembered that she may not spend all her life in the same dairy. She may be sold at just the time she has reached her greatest value. Thus, some cows will not depreciate but will appreciate in value. This is an important fact, which such a formula as would be derived from the above method fails to take into account. The writer knows of some dairies where the practice is to sell cows before they have depreciated to any considerable degree, and a charge based on the above method would be altogether wrong in them. On the other hand, a formula which would represent an appreciation until the cow is say five years old, and then a graduated depreciation might provide the most accurate system of all.

2. *Depreciation on the herd.* By this method, the actual depreciation on a herd for a given year, is estimated as closely as possible, and this amount is divided by the number of cows to get the average depreciation per cow. This amount is arrived at as follows: to the inventory at the beginning of the year, add the price of young stock or new cows entering the herd. From this sum is subtracted the inventory at the end of the year plus the price received for cows sold. This is then divided by the number of cows in the dairy.

It has been objected, that in the case of cows which are sold during the year, an element of profit on such sales will enter into the sum arrived at by this method, as depreciation, and that this will to some extent, invalidate the charge. However, it should be remembered that, in the case of cows purchased, it may just as well be assumed that their price likewise contained an element of profit to the seller, which will offset to the buyer the profit he makes on the cows he sells. At any rate, this difference, in most cases will be so small that the refinement of method necessary to dispose of it, will scarcely be worth while. Depreciation may be computed accurately only after the cow has passed out of the herd, but it is necessary to make some sort of allowance for it each year, and it is believed the herd method is sufficiently accurate for all ordinary purposes.

Allowance should be made for fluctuation in market price, from year to year, as this is not true depreciation or appreciation.

Maintenance of buildings

This factor comprises several subfactors which will be discussed separately.

1. *Interest on investment.* The buildings should be inventoried at the beginning and end of the year. Interest should be charged at the rate the farmer would have to pay if he borrowed the capital invested in the buildings. It is a better business policy to charge interest on the inventory at the beginning than at the end of the year. The principal effect of charging the interest on the basis of the depreciated value at the end of the year, will be to lower the credit the farmer receives for interest. However, this amount of reduction will not be uniform, since buildings differ in their durability. A fairer and more uniform rate of interest will be allowed if it is always charged on the undepreciated value.

2. *Depreciation on buildings.* This varies with the type of the building, but usually runs at from 2 to 4 per cent. It is found by subtracting the inventory at the end of the year from that at the beginning of the year.

3. *Repairs.* A distinction must be kept in mind between repairs, which are of the nature of current expenses and have the effect of making the inventory at the end of the year equal to that at its beginning; and additions which are to be added to the capital invested, and not the current expense account.

4. *Taxes.* The buildings are debited with the actual tax, that is, with the tax paid, at the local rate on the assessed value of the buildings.

5. *Insurance,* is charged at the rate actually paid.

It is not proper to charge for maintenance of buildings, a rate which would cover maintenance of a set of "theoretical buildings," that is, on buildings costing what it is estimated should be sufficient to provide shelter for the herd. But in the case of buildings only part of which are used by the dairy, the charge for maintenance should be made only on that part of the building which is actually used by the dairy.

Maintenance of equipment

This factor includes:

1. Interest on the capital invested in the equipment.

2. Repairs.

3. Depreciation. This factor is generally pretty high, and may run as high as 25 per cent. It may be defined as a charge which is sufficient to maintain the equipment from year to year in equally good repair and to replace articles of equipment as they wear out. Additions to equipment, like additions to buildings, should be added to the capital invested and not to the depreciation account.

4. Tax and insurance on equipment.

These items in the ordinary dairy are so small as to be negligible. However, when an expensive equipment is used, they may assume very noteworthy proportions and must therefore, be taken into consideration.

Cost of bull service

Several different methods have been used in efforts to determine the cost of bull service. None of them have been completely accurate nor eminently satisfactory.

A close approximation to the true cost of bull service is generally the best that can be hoped for. The difficulty of ascertaining the cost of keeping any one of a herd of animals will be readily appreciated when it is borne in mind that farm accounts are poor at best, and the bull is fed out of the same feed bin as the rest of the herd, and cared for by the same men, without any careful account being kept of his feed or care.

The cost of bull service will comprise:

1. Interest on the value of the bull as a herd sire.

2. Depreciation, figured out by practically the same method as for cows, but the period of usefulness will not be likely to be the same as for the cows.

3. Feed.

a. Grain, usually somewhat less than for a cow.

b. Forage, usually slightly more than for a cow.

4. Taxes on his assessed value at the local rate.

5. Expense for shelter.

6. Labor. The bull generally requires more labor than a cow in feeding and caring for him, but this will be offset by the time required to milk the cow.

7. The bull must be charged with his share of the unassignable overhead.

The total cost of keeping the bull is pro-rated among the cows. Only the bulls actually in service should be considered in a study of the cost of milk production. The young bulls, not yet in service, cannot be considered as belonging to the dairy herd.

The device sometimes used, of assuming the cost of bull service to balance with the value of calves at birth, is entirely invalid. The cost per cow for bull service will depend mostly, on the number of cows for which a bull is kept.

Bedding

The cost of bedding is computed in much the same manner as the cost of forage. It is comparatively easy to compute the value of straw used for bedding, because it generally has a market price. The cost of some other materials used for bedding, such as shredded corn fodder, and refuse hay, is not so easy to find, and if their cost cannot be found it becomes necessary to compute their value on the basis of the amount of straw or the usual bedding which they replace.

In the case of some roughages, where the cows eat only a part, and where the rest is used as bedding, it will be necessary to make an approximation of the portion finally used as bedding rather than as forage. This amount is to be charged to the account of bedding rather than that of forage, and at bedding price.

A number of investigations have lumped the expenses for bedding and for forage. A distinction should be made between forage, which, used as feed, stands in some correlation to the amount of milk produced, and bedding which is necessary to the comfort of the cows, and to the production of good manure, whether much or little milk be produced.

Miscellaneous overhead

This will include those minor expenses, which do not vary with production, and which are not of sufficient size to merit consideration as separate factors.

1. Insurance on life of cow.

This factor will be practically constant in all but very exceptional dairies, and may be computed from the mortality tables of live stock insurance companies.

2. Veterinary fees, medicine, disinfectants, etc.

3. Salt, stockfoods, etc.

4. Tax on the assessed value of the cows at the local rate.

5. Water and light.

6. Ice.

7. Cow testing association fees, registration fees, etc.

8. Incidental expenses, as a portion of telephone bills, cost of hiring labor, etc.

VARIABLE EXPENSES

Forage

Forage should be charged to the dairy at farm value when it has thoroughly cured in the mow, that is, at the earliest time at which it could be sold.

In the case of roughage which has no market value to serve as a basis for farm valuation, it may be necessary to compute value as compared to some roughage which does have a market value. Corn fodder may come under this head, but even it has some sort of a market price, as may be found out by visiting a public sale of a farmer's effects.

If interest is to be allowed on the amount of feed kept on hand, this must be charged to the dairy at the farm price at the time the feed is stored, in order to be consistent. The capital on which the farmer may be allowed interest, is that which he tied up by deciding to keep his own feed after the earliest date at which he could have sold it.

The justice of valuing farm crops at the market price (minus cost of delivery) will be appreciated when it is considered that,

in the case of crops grown to sell, the farmer becomes a sort of speculator in his own produce if he does not sell it at the earliest opportunity. Prices may rise later, or they may fall. At any rate, the produce will shrink, and lose weight, which will partly offset any rise in price to come later. The farmer must also provide for, or bear himself, the insurance on loss by fire, storm, and depredation by vermin. Hence, if he keeps his crops, it is at his own risk and his own expense.

On the other hand, if feed is charged to the dairy at the market price at the time it is fed, no interest on its value, nor expense for storage can consistently be charged.

Grain

Grain grown on the farm, should be charged at the farm value at the time of storage, as for forage. Farm value is the market price at the time it is threshed or put in the barn, minus the cost of selling and transporting it to the market.

Concentrates which are purchased, are charged at the price paid, plus the cost of transportation to the farm. In some dairies, feeds may be hauled by the team returning from the creamery or milk station. Here it is not fair to charge the whole expense of hauling either to the feed account or to that of hauling milk, but an allotment of a part of the expense must be made to each.

Silage

Silage should be charged to the dairy at its cost as nearly as that can be found. Since silage is ordinarily grown for dairy feed only, and has no market value in the usual sense, it cannot reasonably be charged at any estimated value. Growing silage must be considered as an adjunct of the dairy business, hence it should be charged at cost.

The same applies to soilage and root crops which are grown solely for dairy feed and not for sale.

Pasturage

Many bulletins have taken what seemed to be the prevailing rate of pasture hire in the neighborhood as the amount to be

charged for this factor. This is hardly a valid method of procedure because, in a bona fide farming neighborhood, there is generally very little "pasturing out," and the rate so computed may not be representative of the actual cost.

The actual cost of pasture may be found by adding together the following factors:

1. Interest on the value of the pastured land, at the prevailing rate in the neighborhood.
2. The cost of keeping up the fences of the pastures.
3. Cost of reseeding, mowing, etc.
4. Fertilizers and top dressing applied to the pastures.
5. Taxes on the pasture lands at the local rate.

Labor

Many and varied methods have been employed in computing the cost of labor, but the simplest and the one which measures individual differences most accurately will prove the most reliable and satisfactory in the long run, and the most dependable in making comparison between different dairies or groups of dairies.

The actual cost is to be found by adding up cash wages, and value of board, and other perquisites which the laborer receives. This sum is divided by the number of hours the man worked during the month or week, as the case might be, to find the cost per hour. No greater period of time than the month should be used in computing the cost per hour of labor, because otherwise an error would enter in from the seasonal variation in wages.

Horse labor

The cost of keeping a horse on a given farm for a year, must serve as the basis for finding the cost of horse labor per hour, the cost per year being divided by the number of hours of labor the horse performed during the year to find the cost per hour.

It is necessary to use the cost of keeping the horse a year instead of a month, as in the case of man labor, because of the wide seasonal variation in the number of hours of horse labor performed per day on the average farm. If it is necessary to keep the horse during the winter, when he does not work, in order to

get his services during the summer, it is only fair that the enterprises on which the horse works during the summer should bear a share of the expense of keeping him during the winter.

Cost of hauling milk

In a study of this sort, there are two possible alternatives.

1. To find the cost of producing milk on the farm and leave out of account the cost of hauling, or in other words, to find a farm cost.

2. To find the cost of the milk as delivered at the creamery or milk station.

Since the latter is the actual cost to the farmer, and he seldom sells his milk on the farm but is required to deliver it to some point at a distance, it will usually be the one used. The following method of computation may be used;

1. Find the actual number of hours spent in hauling the milk and in returning to the farm, as nearly as possible.

2. Multiply this number of hours by the cost per hour of the labor of the man who drove the milk wagon.

3. Multiply the number of hours by the cost per hour of the horses labor.

4. Add a reasonable amount for depreciation on the value of the wagon and harness.

It may be desired to know the cost of hauling a can of given capacity, per mile. This may be easily found by multiplying the number of cans of milk hauled by the number of miles to the milk station, and dividing by this product the total cost for hauling.

Incidentals

This includes those items which vary with the amount of milk produced, but are not of sufficient size to merit individual treatment. Such items are:

1. Feed grinding.

2. Storage of feed.

3. Interest on funds usually kept on hand to cover operating expenses.

4. Interest on the capital tied up in stored feeds, including both those bought and those grown on the farm. If this interest is allowed the feed grown on the farm must be charged to the dairy at the farm price at the time it is put in the barn. And if this charge is not allowed the feed will be charged at the price when it is fed. The price at the time the feed is fed, will include, theoretically at least, a charge for storage, and for interest. The charge for storage, if it is made, may be either incorporated in the price charged the dairy for the feed or, perhaps better, made separately, while the interest is added to the general charge for interest.

CREDITS

Credits for milk

Credit for milk will comprise the annual income from milk delivered at the milk station or creamery. If it is desired to compare prices received in different dairies, it will be necessary to reduce the prices to some comparable status. Some dairies deliver their milk to creameries, and so have no freight to pay. Others ship to distant dealers, and the price they receive must be reduced by the amount of the freight before it can be compared to that received by the other class.

All milk used at the farm for home consumption, and for raising calves, after the earliest time at which the calves could be weaned, comes under this account. Skim milk in dairies where the milk is skimmed on the farm, and only the cream or butter sold as a direct product of the dairy, is also to be included under the heading of credit for milk. This skim milk is usually marketed indirectly by feeding it to hogs, and is thus a considerable source of income to the farm.

Credit for calf

The dairy should be credited with the value of the calf at three days of age, the time at which it could be separated from the cow.

For the purposes of the ordinary cost of production study, calves of similar size, conformation and vigor should be valued

at the same rates, whether they are grades or pure bred. To introduce the factor of additional value inherent in the fact of registry would greatly complicate the study, and for the purposes of such studies, this peculiarity of pure bred calves may be considered as a return on the corresponding value of their dams. This device of valuing pure bred calves at the same rate as grades, also conforms to the system given above for valuing the cows.

Credit for manure

Manure should be valued at a rate which would enable the farmer to buy equivalent amounts of other fertilizers containing the same elements of plant food. Allowance should also be made for the value of manure as improving the physical and biological properties of the soil. It will usually be necessary to adopt a uniform rate per ton. It will seldom be possible to go into such refinements of method as to place separate valuations on the manure on different farms, although such differences are known to exist as a result of different methods of handling manure, excessive leaching on some farms, etc.

The rate adopted per ton should be determined by the usual or average condition of manure in the neighborhood, when it is hauled to the fields.

Other credits

In most dairies there will be a few other credits than for milk, calves, and for manure. These are such as for hides, bags, etc. They are not likely to form a very important factor but they should not be lost sight of.

In the dairy business, the milk is the foremost aim of the business, and the other products are merely incidental to the production of milk. Hence, in finding the cost of the milk, the sum of all other credits are first subtracted from the total cost of production, and the remaining, or net cost is taken to be the cost of the milk.

PRESIDENTIAL ADDRESS¹

C. H. ECKLES

St. Paul, Minnesota

This is the sixteenth meeting of the American Dairy Science Association. I believe this organization has performed a distinct service to the dairy industry of the United States.

The preamble of our constitution states that "The object of the association shall be to advance the general welfare of the dairy industry, especially, by the improvement of dairy instruction, the stimulation of scientific research, in all phases of the subject, and by improvement in methods of conducting extension work." I believe the organization has in a generous measure served this purpose in the past.

In my judgment there has been marked advances in every line of the several activities in which our members are engaged. Instruction in dairying was begun only about thirty years ago. At that time no one had any experience in teaching the subject. The inevitable result was that considerable time was required to get the subject into pedagogical form so it could be taught. This accounts for much of the inferior teaching we all know has been done along this line in the past and for the lack of uniformity which has been so noticeable in the past in the arrangement of our courses of instruction.

One of the marked advances during recent years has been the improvement in the college instruction along dairy lines resulting from a better understanding of what should be taught and how the teaching should be carried on. This improvement is certainly due to a considerable extent to the work of this organization, especially through the committee reports and discussions at our annual meetings. I feel confident that the instruction in

¹ Delivered at the opening session of the Sixteenth Annual Meeting of the American Dairy Science Association at St. Paul, Minnesota, October 11, 1921.

dairying has been improved in every institution in the country that has had a representative at one or more of our annual conferences. Not only does the information derived from our discussions and from conversations with those from other institutions widen our knowledge regarding methods of instruction but, perhaps greater than all, it supplies a stimulation for doing things better that cannot be fairly measured.

The college instructor in dairying has some serious problems still before him. With the rapid increase in attendance at all agricultural colleges the question of how to handle large numbers of students in laboratory work is becoming a serious one to practically every instructor in dairy work. Efficient instruction in dairy work requires abundant and expensive equipment, together with a large force of instructors in proportion to the students. It is recognized that the function of a dairy department in all agricultural colleges is not alone to give instruction in any limited field of dairy manufactures. No dairy department is complete without provision for instruction in all that pertains to dairy cattle, bacteriology of milk including sanitary market milk, and the manufacture of butter, cheese, condensed and powdered milk, and ice cream. Another problem is how much purely practical work a student in dairying should be given, and what part of his time should be devoted to securing a scientific training fundamental to the work for which he is preparing himself. In other words, is it the duty of a Dairy Department to turn out men qualified to immediately take hold of large business enterprises or is it our duty to give men a good fundamental training and expect they will gain most of their practical experience after leaving the institution?

The tendency of the public is to expect that our graduates shall be able immediately after leaving school to carry large responsibilities and as instructors we perhaps encourage this idea unduly. Why should we not look upon instruction in agriculture more as is done with the technical instruction in engineering? It is not expected that an engineer is competent to assume large responsibility immediately upon graduation, but that he has the fundamental training which makes it possible for him within a short time to carry such responsibility.

Another important question is that of more and better qualified men for college and experiment station positions. We are not turning out enough thoroughly prepared men. There should be more graduate students in dairy husbandry than is the case at present. The time is already here when a man to fill a first class college position and rank with other men with whom he is associated must have a good graduate course. I am glad to say that graduate students are now enrolled in some of our institutions working for a doctor's degree along dairy lines. I believe it is our duty to encourage this kind of work in the stronger institutions. I do not in any way minimize the importance of practical instruction, but at the same time I feel it is even more to the credit of the institution and more service to the state to turn out a few thoroughly qualified men who become leaders in their lines than it is to over emphasize the importance of turning out a graduate who can immediately turn his hand to practical work but whose training has been inadequate to enable him to do real constructive work of permanent value.

Another of the objects of this organization should be to stimulate research work. A few men in college work may gain prestige and be of great service to the state on account of executive ability, others through special attainments in the way of teaching, still others through their ability to disseminate information become leaders in extension work; for others of us perhaps the best opportunity to do something that will be of real value to the world at large and at the same time bring us some credit lies along the line of scientific investigation.

The research side of dairy husbandry work has certainly received great stimulation within recent years. There is unquestionably far more good work of this kind under way at the present time than has ever been the case at any one time in the past. I do not believe it is boasting to say that this organization has also been a considerable factor in bringing about this improvement. Anyone familiar with research work in dairy lines knows that in the past most of the best work has been done by men who were trained especially in some certain scientific line, as, for example, bacteriology or chemistry, and were not

primarily interested in technical dairying, and the same will probably be the case in the future. The man filling a dairy position has such varied responsibilities to carry, including the business management of a considerable business enterprise, teaching regular classes, and carrying on extension work, that he has little time for investigation. Further, his subject involves several distinct scientific lines, which adds to the difficulties from a research point of view.

There is no reason why the head of the dairy department in a college or a man filling a responsible government position should not, however, be a real leader or director of the most valuable and fundamental scientific work that may be done along dairy lines. He must not make the mistake of trying to do it alone. The time is past when it is possible for any one man to attack a problem of any size alone when it involves as widely separate lines as the feeding and management of the animal and the application of chemistry and bacteriology. Such investigation must be carried on largely by the association of men familiar with the various phases of the work. Practically nothing can be done in the way of research work relating to either dairy products or dairy cattle without the assistance of a chemist. Whenever research work involves the animal then we will sooner or later need the services not alone of a chemist but of a physiological chemist. It seems probable that the most important results in connection with experiments involving animals within the next few years will be brought about by applying the principles of physiology and chemistry together with the knowledge of the expert in handling animals.

Dairy husbandry is the application of several sciences to certain practical lines and for this reason a man directing such work should have a rather broad training without being necessarily a specialist in any one. When a chemist alone undertakes to carry on an investigation with animals he is apt to overlook some of the essential points in treatment of the animals used and he is not in a position to know the problems that need solution from a practical standpoint. On the other hand one filling a responsible position along dairy lines does not have as a rule the intimate

knowledge of chemistry and especially of physiological chemistry necessary to properly carry on a research to a good advantage. The man in this position, however, has an opportunity to know the problems that the practical man wants solved and he should combine with this the technical knowledge of the management of the animals or the manufacture of the dairy products, as the case may be. The plan that must be followed in the future if we gain much headway is to work in groups. One of the group must have a broad general knowledge and be familiar with the practical side of the problem. Associated with him must be competent chemists, physiologists and bacteriologists according to the problem at hand.

The Dairy Division of the United States Department of Agriculture continues to fulfil its position of leadership. The United States government has always been the real leader in popular education in this country. The state universities were founded as the result of an act of Congress. Our agricultural colleges were established by the same body. Agricultural research was begun by the Hatch act establishing the experiment stations and later further stimulated by the Adams act. The Dairy Division through its wide dissemination of knowledge and its leadership in improving market milk, developing better methods in creamery work, assisting in the introduction of modern methods in the South, and taking the lead in encouraging fundamental research, is doing constructive work, the influence of which will still be felt in generations to come.

The object of the Dairy Science Association is not the dissemination of information regarding improved methods which is the function of other agencies. Our business is rather that of considering, as those in positions of responsibility, how we can best do our work, how we can best carry knowledge already known, to the public in general by extension work, and to our students by class work instruction, and how we can so direct our efforts as to make the most rapid advance in the acquisition of new knowledge.

The agricultural colleges and the United States Department of Agriculture have already obtained a position of leadership in

American agriculture. Each member of this Association bears a part of this responsibility. To a large extent we are depended upon by the industry which we represent to lead them in the right direction. We are either leading the people right or we are leading them wrong. The dairy industry already represents a value of nearly two billion dollars each year. Only two or three products of the farm exceed it in value. We also realize great responsibility of improving the conditions under which this enormous amount of human food is put upon the market to the end that it may not endanger health. The future development of the industry in which we are interested is to no small extent under our influence. It is our duty to help place the agriculture of America on a permanent basis.

If time would permit it might be profitable to enumerate some of the past achievements of the American Dairy Science Association. Important as these are there remain so many possibilities of usefulness before us that our attention should be directed mainly toward the future and its problems.

MEMBERSHIP

Our membership is now the largest in the history of the Association. Through the activities of our membership committee, a report from which will be presented for your consideration, over fifty new members have been added within the past two months. In making this campaign for enlarging our membership the aim has been to fully maintain the standards of our association as prescribed by our constitution. The question of how wide the door of membership should be opened, you will recall, has been the cause of considerable difference of opinion in the past. Membership in this organization was originally limited to men in college experiment station and extension work and in the United States Department of Agriculture. When our present constitution was adopted the organization was broadened somewhat and became in reality a professional association. This is not a business association, but a professional group corresponding somewhat to the bar association, medical associations and various chemical societies. Undoubtedly the eligibility re-

quirement as it now stands in the constitution should be clarified somewhat as it is too indefinite. A report of the committee on constitution and by-laws will be presented dealing with this subject.

I am strongly of the opinion that the door of membership is now open wide enough to admit all who will add strength to our Association. I see no special advantage in changing our basis of membership merely to increase our numbers. However, all who are properly qualified should be offered membership. I desire also to recommend a change in our method of increasing membership. At present we wait for candidates to apply and we urge them to apply for membership. It is decidedly unpleasant to reject a candidate who does apply under these conditions and presumably some do not apply who are eligible and who would like to be members, on account of uncertainty as to their eligibility.

A much more dignified and more business like plan would be to elect men to membership without formal application on their part, leaving it to them to accept the membership offered them or decline it as they see fit. To put our election of members on this basis would require some little activity on the part of the regular membership, to be sure, since it would devolve upon them to nominate members. It would also add somewhat to the burdens of the secretary. A plan of this kind carried out with the full coöperation of the members would bring in most of those who should be members and if the eligibility for membership is properly safeguarded would make election to membership mean much more than it does under our present procedure.

JOURNAL

The Journal of Dairy Science is a publication of which we may well be proud. I wish to commend the work of the editor most highly. Our Association is greatly indebted to Professor Frandsen for his services in this position. If our organization had never done anything else but establish this journal our efforts would be well repaid.

SECTION ORGANIZATION

In accordance with the constitution adopted two years ago the meeting held in October, 1920, took the first steps towards the organization of sections. Three were authorized by the executive committee as follows: (1) Dairy production; (2) dairy manufactures; (3) official testing. At the annual meeting of the Association of Extension Workers in Dairying it was voted to join the Dairy Science Association, constituting a dairy extension section. I am sure all welcome the addition of this important group as a section of the Dairy Science Association.

The section plan of meeting makes it possible for each important interest to have its full share of time for the presentation of reports and papers. Much remains to be done in the way of completing the details of the organization and getting a program for the future under way. Steps should be taken to classify the membership according to the section with which they wish to especially affiliate. Presumably many members will desire to retain their relations with more than one section and I see no objection to such an arrangement. For the administration of the work of both the sections and of the general association such lists are essential.

PROGRAM

In my judgment the most important step before the Association is the formulation of a more definite program for its activities in the future. The importance of this was clearly brought to our attention by the chairman of the dairy manufactures section in a recent number of the *Journal*. I trust the committee on organization for the sections will have something to present along this line.

Closely connected with this matter of a general program of work is that of the character of a program to be given at our annual meeting. I am not entirely satisfied with the character of the programs we have been accustomed to have on these occasions. Possibly we have depended too much, for one thing, upon committee reports. A certain number of committees are a very essential part of our organization and some construction

work of a high character has been done by them in the past. Standing committees to deal with certain subjects should unquestionably be maintained and most of these will do the best work if the membership remains fairly stable from year to year. Among committees of this type I would place those dealing with standards, score cards and judging contests. Committees of this class should not be expected necessarily to report at every meeting. They should remain on guard to study any situation that may develop and to report as often as it seems advisable.

There are committees of another type that should be relieved when the specific purpose for which they were appointed has been accomplished and their report has been submitted. In this class I would place such committees as those dealing with courses of study and feeding standards. If such committees are continued the membership should be changed from year to year. When a man has once given his ideas along a certain line he should not be called upon the next year to give to report again on the same subject. I am of the opinion that in many cases better results would be had if an individual member was asked to prepare a report or paper upon subject in place of appointing a committee of several members. This statement is clearly limited to subjects of a certain class. For example, one member could and probably would prepare a better report upon graduate instruction in dairy husbandry than a committee could. If a subject of this kind is in the hands of a committee the report, if it is forthcoming, must of necessity be almost exclusively the work of some one member of that committee. On the other hand where the subject matter is of another type combined action of a committee is clearly essential, for example in the case of standards and official testing.

I am not pleading for the abolishment of all committees but that they be rearranged with a view to deciding what standing committees are needed and what should be their responsibilities. All committee reports should be prepared in proper form for publication in the *Journal* and published later if deemed advisable by the board of editors. If the report is too long a résumé only should be presented at the session of the association.

Most other societies of a similar nature present a program either in the nature of scientific papers or addresses and reports by individual members. A common practice at scientific meetings is to have abstracts read of papers to be published in the journal of the society later. I believe this feature could be added to our program with advantage. I have no thought that it would be desirable to make the entire program of this character but to add it as one feature. My suggestion is that the program should be a combination of (a) reports of standing committees, (b) papers or reports by individuals who have been asked to make reports on particular subjects, (c) abstracts of scientific papers by members—these to be limited to five or eight minutes each with the understanding that the full paper be submitted for publication in the *Journal* later. It would seem logical to place the responsibility for the programs of the sections upon the officials of the sections, leaving the general officers the responsibility for the general program. Probably there should be a provision in the by-laws covering this point, although such provision is not found in the by-laws at present it was assumed to be the proper procedure and the responsibility for the assignment of the programs to be given by the sections today and tomorrow was definitely given to the chairman of the several section.

ORGANIZATIONS OF DIVISIONS

The constitution makes provision for the organization of divisions of the Association based upon geographical considerations. There has been for several years an organization including a group of men engaged in college, experiment station and extension work on the Pacific coast. The suggestion has been made to members of this group that their organization become a western section of the Dairy Science Association. I believe the matter has not been passed upon as yet by that group. A committee representing a similar organization in the eastern states will present a request today that an eastern division of the Association be authorized. This is entirely in accordance with our constitution and is a most desirable arrangement. While the

constitution places the authority for the authorization of sections and divisions with the executive committee, since the matter comes up for action at the time of our annual meeting it will be brought before the Association for action.

RECOMMENDATION OF THE EXECUTIVE COMMITTEE

The report of the secretary-treasurer shows the Association to be in a healthy condition financially. The income for the coming calendar year available for the uses of the association is estimated at \$625, which in addition to our present balance on hand will make possible a forward step in the affairs of the organization.

The Association by action of its executive committee has appropriated since the *Journal* was established \$100 yearly for the expense of the office of the editor. This sum falls far short even of paying clerical help and postage. This means the business of our association has been financed in part by the institution and later the company with which our editor has been associated. In addition our editor has donated his services. The only compensation he has ever asked has been the support of his colleagues and the proper appreciation of his efforts. In view of our present improved financial condition we recommend that beginning with the next calendar year the sum of \$300 annually be appropriated for the use of the editor of the *Journal* in paying office expenses and advancing the interest of the *Journal* in whatever way he may find desirable.

The position of secretary calls for a large amount of detailed work as well as the contribution of much constructive thinking. The plans of our organization and our enlarged membership will result in even greater demands on the time of the secretary in the future. We have reached the point where the secretary should receive some compensation for his services and should have sufficient funds to pay the necessary office expenses. We recommend that the secretary receive a compensation of \$100 for each calendar year, with an appropriation of \$100 for office expenses with the understanding that additional sums may be used with the approval of the executive committee.

This action as recommended we feel is only a step in the direction in which we shall advance year by year until some time in the future we shall have a full time secretary and an editor receiving a salary in keeping with the position our *Journal* will have reached by that time.

Behind the activities we expect to have a membership adequate to properly support the organization and one whose leadership in the advancement of scientific dairy husbandry shall be unquestioned.

ANNUAL MEETING OF THE AMERICAN DAIRY SCIENCE ASSOCIATION¹

GENERAL SESSION

The sixteenth annual meeting of the American Dairy Science Association was held in connection with the National Dairy Show at the Twin Cities, October 11, 1921. The meeting opened with an address by President Eckles, following which Professor Regan reported on the question of an eastern section of the Dairy Science Association. It was proposed that this section hold their annual meeting in connection with the Eastern States Exposition. It was moved by Dr. Harding that it be the sense of this meeting that we favor the formation of an eastern section, the geographical location of which shall be determined by the executive committee. This motion was carried.

A letter from Professor Hastings was read regarding the representation of our Association on the National Research Council. Moved by Professor Stocking that this matter be taken up by the executive committee with the Council, looking toward a representation of this organization. Motion carried.

The report of the secretary showed that the Association is in very good financial condition.

After a brief discussion of the secretary's report, it was the opinion of the Association that some funds, placed at the disposal of the secretary and the editor, would enable them to more rapidly advance the interests of the Association.

It was moved by Professor Davis that the recommendation of the executive committee be approved and the following budget be adopted for the year of 1922:

Editor, salary and clerical help.....	\$300.00
Salary of secretary.....	100.00
Expenses and clerical help of secretary.....	100.00
	<hr/>
	\$500.00

¹ Credit is due President C. H. Eckles and Secretary N. W. Hepburn for assembling the data for this report.—*Editor*.

Motion carried.

Professor Lockwood moved that we commend the Department of Agriculture for their very fine exhibit at the dairy show this year. The motion carried.

Moved by Professor Gamble that we send an expression of our appreciation to those who have furnished cups and medals in connection with our student judging contest held at the show. Motion carried.

PRESENTATION OF REPORTS

The attention of the session was next turned toward committees reporting to the general session.

REPORT OF THE MEMBERSHIP COMMITTEE

J. A. Gamble, Maryland, Chairman

(An abstract of this report is published on page 156.)

Following this report it was moved by Professor Hunziker that the membership be classified into sections and that each member be furnished with a list of the membership showing complete classification. Motion carried.

REPORT OF COMMITTEE ON CONSTITUTION AND BY-LAWS

Prof. M. Mortensen, Iowa, Chairman

The committee recommended that article 3 of the Constitution, defining membership, be amended to read as follows:

Article 3. Membership shall be of one kind only; namely, Active.

Article 4. The following are eligible for election: Any person who is formally announced by an Agricultural College, or Experiment Station, or by the Dairy Division of the United States or Canadian Departments of Agriculture as an instructor, extension worker, investigator or administrative officer connected with the dairy industry, or anyone filling a position of responsibility connected with the dairy industry, and who has had a college or university training in technical science, or anyone filling a responsible position in the industry of a professional character requiring a technical knowledge of dairying of a high order.

The following additions to the By-Laws were recommended:

Article 6. A committee of five on elections shall be appointed by the president. Said committee shall make a report at the annual meeting in which the names of

two members shall be suggested for each office to be filled by mail ballot under the provision of the constitution. In addition thereto, a group of five or more members may suggest other names for consideration, but such requests shall be in the hands of the secretary of the association before November 1st.

Article 7. In conducting mail ballots only those ballots shall be counted which are received by the secretary within four weeks from the date upon which the ballots were mailed by the secretary.

Article 8. Section Meetings. The time and place of meeting for the Sections of the Association shall be fixed by the Executive Committee. All other arrangements regarding the section meetings shall be made by the officers of the section.

These recommendations were adopted. The constitution and by-laws as revised by this action are printed in full on pages 194-197.

REPORT OF THE COMMITTEE ON BACTERIOLOGICAL METHODS FOR MARKET MILK

Dr. R. S. Breed, New York, Chairman

An informal report was presented by the chairman calling attention to the publication of the third edition of the Standard Methods for the Bacteriological Examination of Milk in January, 1921. Copies of this report are obtainable from the Secretary of the American Public Health Association, 370 Seventh Avenue, New York City. The report as published is a joint report of committees representing the American Public Health Association, the American Dairy Science Association, the International Dairy and Milk Inspectors Association, and members of committees from the Society of American Bacteriologists and the American Association of Medical Milk Commissions.

It was also stated that in New York State, where the matter of paying for milk as delivered by dairyman on the basis of the bacterial count, has made rapid strides, a bill is to be introduced at the coming session of the legislature establishing a control over the making of bacterial counts where these are used as a basis for payment. The essential part of this control is that analysts must submit to examination in order to secure a license, that graduated glassware is to be examined for accuracy by the State and that the methods used in taking samples and in carrying out the analyses are subject to review by state authorities.

REPORT OF COMMITTEE ON DAIRY SHORT COURSE INSTRUCTION

Prof. J. R. Keithley, Minnesota, Chairman

(This report is published in detail on page 160.)

The meeting adjourned for lunch, the President announcing that the membership would convene by sections for the afternoon session.

PROCEEDINGS OF SECTION I, DAIRY PRODUCTION

Meeting called to order by Chairman Regan. Secretary absent.

P. S. Williams was appointed temporary secretary.

The possibilities of a new dairy cattle score card were discussed. The opinion of members present seemed to be in favor of a revised score card. It was moved and seconded that a committee be appointed to study the score card and make suggestions at the next meeting. Reports of standing committees were next called for.

Report of the committee on methods of conducting student judging contests. W. W. Swett, Chairman

Mr. W. W. Swett read report. Moved and seconded that the report of the committee be accepted. It was the opinion of some that the name of the official judge of each breed for the contest be published in the catalogue and that the same judge be used for the contest as for the show.

Mr. Borland brought up the matter of an individual judge for the contest and stated that the official judge in a certain breed was not upheld in his judgment by 40 out of 45 contestants. He was in favor of a three judge system. The matter of appearance of cows at time of judging being different when officially judged and when judged by the contestants was discussed and it was thought advisable to have the judge see the cows at the same time as contestants.

Professor Rayburn suggested that the judge select his own cattle for the contest. It seemed to be the opinion of several men that the judge should select the cattle.

H. P. Davis was in favor of having 5 or 6 animals in each class, at least in female classes and reasons only be used in classes where prizes are offered.

Professor O. E. Reed suggested that a committee be selected from the group of coaches, 2 or 3 men to judge each breed. This committee to be selected by a committee of the American Dairy Science Association for the contest.

The report of the committee was accepted by unanimous vote of the section.

*Report of committee on organization. W. W. Swett, Missouri,
Chairman*

A constitution and (by-laws) were drawn up by the committee and read at the meeting. A suggestion was made by Ragsdale to have meetings of sections at separate times. The question of limiting of members was discussed. Ragsdale proposed that at certain times all members should not be allowed to vote.

H. P. Davis moved that article 3 concerning membership as read by committee be accepted. Motion seconded and carried.

The reading of article concerning officers shall read chairman rather than president.

Mr. Eckles suggested that few standing committees be appointed. The report of the organization committee was accepted and Mr. Dice was appointed as a committee on dairy score card to report next year.

Officers elected were: W. M. Regan, New Jersey, chairman; A. C. Ragsdale, Missouri, vice-chairman; E. L. Anthony, West Virginia, secretary.

Adjournment.

(Signed) PAUL S. WILLIAMS,
Temporary Secretary.

PROCEEDINGS OF SECTION II, DAIRY MANUFACTURES

Session called to order at 2:00 p.m., in Dairy Hall, University of Minnesota. In the absence of Chairman Potts, Professor A. W. Rudnick presided. The first order of business was the call for the report on "Organization."

*Report of committee on organization. O. F. Hunziker, Chicago,
Chairman*

Your committee on organization of the section of dairy manufactures beg to submit the following report:

1. *Membership.* Your committee recommends that any member of this Association be eligible to the membership of the section of dairy manufactures, and that each Association member desiring to become a member of this section be requested to so register by filling out a card of membership furnished by the Secretary of this Association.

2. *Standing committees.* It is the sense of your committee that there is danger of confining the activities of this section too exclusively to committee work and committee reports. This, we fear, may disadvantageously affect the attractiveness of our sessions and the service it is our purpose to render the dairy industry.

We feel that the purpose and effectiveness of our work demand that the number of subjects assigned to committees be restricted to the minimum and that committees only be appointed on subjects for the adequate consideration of which committee work is indispensable.

We, therefore, beg to recommend that the committees for the Section of Dairy Manufactures for the ensuing year be confined to the following standing committees:

Committee on organization

Committee on legal standards and score cards for dairy products

Committee on official methods for testing dairy products

Committee on dairy products judging contests

3. *Program.* Your committee beg to further recommend that the sectional leader shall appoint individuals to present a résumé covering the current situation and other matters of vital interest regarding the diverse phases of their respective branch of the industry; these individuals to be appointed immediately after the regular annual meeting. These résumés or reports should

form a most valuable part in the program of the annual meeting of the succeeding year.

R. S. BREED,
J. H. FRANDSEN,
J. A. GAMBLE,
O. F. HUNZIKER,
Committee.

Report of committee on legal standards and score cards for dairy products. J. H. Frandsen, Chairman

(This report is published on page 164.)

Moved that the legal standards committee be instructed to investigate the weight and total solids standard for ice cream. Motion carried.

Report of committee on dairy products judging contest. Prof. A. W. Rudnick, Chairman

(This report is published on page 168.)

Following this report it was moved by Professor Mortensen that a special committee with Professor Gamble as chairman, be appointed to study the rules and regulations pertaining to the judging of milk. Motion carried.

Professor Bouska made a motion favoring the appointment of a committee for the purpose of standardizing the terms used in criticizing the characteristics of butter in reference to judging the same. Motion carried.

Professor Mortensen moved that a list of ten or twelve judges of national reputation for each of the three dairy products be given to General Manager Skinner and that judges for dairy products in the student contests be selected from this list. Motion carried.

Moved that not more than one judge can hold over from one year to the other. Carried.

Report of committee on official methods for testing milk and cream for butter fat. O. F. Hunziker, Chicago, Chairman

(This report is published on page 175.)

The following were elected officers of the Manufactures Section for the ensuing year: L. A. Rogers, Washington, D. C., president; C. L. Roadhouse, California, vice-president; H. A. Ruehe, Illinois, secretary.

Adjournment.

A. W. RUDNICK,
Secretary.

PROCEEDINGS OF SECTION IV, OFFICIAL TESTING

The following men were present at the meeting: E. L. Anthony, West Virginia; R. B. Becker, Kansas; P. M. Brandt, Oregon; H. P. Davis, Nebraska; J. R. Dice, North Dakota; C. H. Eckles, Minnesota; L. H. Fairchild, Indiana; C. R. Gearhart, Kansas; Roy T. Harris, Wisconsin; L. W. Morley, Pennsylvania; W. E. Peterson, Minnesota; A. C. Ragsdale, Missouri; Wm. J. Regan, New Jersey; Chas. W. Turner, Missouri; P. S. Williams, Pennsylvania; H. H. Wing, New York; E. G. Woodward, Washington; C. E. Wylie, Tennessee.

Voting membership in the section is limited to heads of Dairy Departments, and men actively in charge of Official Testing work. Others especially interested are invited to attend meetings, but have not the privilege of voting. Voting is done by states, each state having but one vote.

Meeting called to order by chairman H. P. Davis.

In the absence of Secretary M. H. Fohrman, the chairman appointed R. B. Becker (Kansas) as temporary secretary.

Moved by A. C. Ragsdale and seconded that the reading of the report of the last meeting be dispensed with. Motion carried.

Report of the committee on relation to breed associations. E. G. Woodward, Washington, Chairman

The report of this committee has been divided into three sub-committee reports as follows:

1. Administration, Roy T. Harris and W. W. Yapp.
 2. Uniform rules, H. N. Colman and J. B. Fitch.
 3. Financing test work, Wm. J. Regan and G. C. White.
- (These reports are found on page 183.)

1. Report of the sub-committee on administration of official testing work within the United States. Roy T. Harris, Wisconsin, Chairman

(See page 183)

It was decided to read reports section by section, making amendments, and voting on the completed reports.

Discussion. H. H. Wing approves the adoption of uniform report forms, and elimination of all material except scattered or previous milkings.

H. P. Davis and C. W. Turner suggested that a full exhibit of forms used by the different institutions should be collected for exhibit at the next annual meeting.

Moved by E. L. Anthony that the secretary gather the forms used by the institutions, and prepare an exhibit for the committee on relation to breed associations, and that they prepare tentative general forms to submit to the different institutions. Motion passed.

Moved and seconded that report on "administration" be adopted as approved. Motion passed.

2. Report of the sub-committee on uniform rules for the conduct of official records of dairy cows. H. N. Colman, Oregon, Chairman

(See page 184)

Report of sub-committee on uniform rules read by P. M. Brandt, substituting for H. N. Colman. Rules read by section. Voting was conducted by states.

Speech of welcome made by the chairman, H. P. Davis, to the representatives of the breed associations who came at this time. These were:

Secretary and Mrs. Malcolm H. Gardner, Holstein-Friesian Association of America.

Secretary Wm. H. Caldwell, American Guernsey Cattle Club.

Secretary C. L. Burlingham, Ayrshire Breeders Association.

Secretary R. M. Gow, American Jersey Cattle Club.

Chief of registry of merit O. H. Baker, American Jersey Cattle Club.

Secretary Caldwell said in substance: The American Guernsey Cattle Club wishes to retain absolute control of the conditions under which a cow can enter the Advanced Registry. The Club asks that the owner's reports be absolutely accurate and honest in reporting the milk and feed record. The Club asks that the state institutions, as a third disinterested party, report the official tests. The Club desires the close coöperation of the state institutions in maintaining the authenticity of the advanced registry work.

Secretary R. M. Gow spoke about as follows: The American Jersey Cattle Club appreciates the help the colleges have been in management of tests. Any rules which the colleges feel would make the records still more accurate, will be supported by the Club. The American Jersey Cattle Club recognizes any rules made by the state institutions supervising tests which are practical and proper to make in safeguarding the register of merit work.

O. H. Baker, Chief of Registry of Merit Department, spoke about as follows: The American Jersey Cattle Club has certain fundamental rules which are required, and in addition will support any additional rules of any state institution to safeguard the records. The right is reserved to chart apparently abnormal production records by the "graph system."

Secretary Gardner discussed the methods of using figures reported to the third decimal place in computing records. He asked that the states select supervisors who will not be influenced by environment or by flattery. The need is for supervisors who will give an impartial report of the production of cows on test. The graph system, as used by the Holstein-Friesian Association of America in charting apparently abnormal production, was explained.

Secretary C. L. Burlingham found the same problems in handling breed work with the Ayrshire cattle that have been found with the other breeds. He praised the organization of the American Dairy Science Association in trying to bring about uniformity and accuracy in handling official testing work.

The addresses of the breed representatives were followed by a full discussion of problems affecting both the state institutions and the breed associations, and a much better understanding of them obtained.

Mrs. M. H. Gardner spoke especially of the conditions which test supervisors are called upon to meet, and praised the integrity of these men in maintaining a high standard of excellence in their testing work.

3. Report of sub-committee on methods of financing official testing.
Wm. J. Regan, New Jersey, Chairman

(See page 189)

Full discussion of financing problems was taken up, including consideration of methods of providing funds for financing official testing work. Consideration was given to (1) breed associations handling finance of tests; (2) breed associations providing money to establish rotating funds at the several institutions; (3) that breed associations act with breeders in each state, asking the legislature to provide a rotating fund; (4) that each institution, by fees charged to each breeder, gradually build up its own revolving fund.

Moved by A. C. Ragsdale that the recommendations or suggestions of the Official Test section of the American Dairy Science Association be presented in writing to the breed secretaries. Motion carried.

Report by W. E. Peterson (Minnesota), Roy T. Harris (Wisconsin) and R. B. Becker (Kansas) concerning the value of a preliminary milking in getting a representative production during the two-day test period, upon which semi-official records are largely based. It was stated that a full report of experimental work conducted by the Kansas Experiment Station will be reported in the JOURNAL OF DAIRY SCIENCE.

C. H. Eckles reported on the attitude of breeders and herdsmen toward the work of the supervisors.

Moved by W. E. Peterson that a rising vote of thanks be extended to the representatives of the breed associations for attending the meeting, and taking part in the discussion of problems affecting official testing work. Motion carried.

Report of sub-committee on "uniform rules" was resumed.

Moved by P. M. Brandt and seconded, that article VI be taken from the table, and considered for adoption. Motion carried.

Article VI adopted.

A. C. Ragsdale recommended that section III be offered as suggestions to bring before the breed associations.

Moved by Wm. J. Regan, seconded by E. G. Woodward, that report of sub-committee on uniform rules be adopted. Motion carried.

Moved by E. G. Woodward and seconded, that report of sub-committee in financing test work be adopted.

Discussion: Report does not cover question of revolving fund to finance official testing work.

Motion carried.

Moved by A. C. Ragsdale and seconded that chairman of committee on relation to breed associations submit copies of these rules to the different institutions supervising tests, and that the recommendations be presented to the officials of the dairy cattle breed associations. Motion carried.

Moved by L. H. Fairchild that chairman of committee on relation to breed associations arrange for a meeting of officers of the breed associations with this committee at an early date. Motion seconded and carried.

Report of committee on by-laws for the official testing section of the American Dairy Science Association, presented by E. G. Woodward.

Moved and seconded that report of committee on by-laws be accepted as finally approve Motion carried.

Moved by Wm. J. Regan that L. H. Fairchild be appointed a committee of one to prepare and report at the next annual meeting on the "composite test." Motion carried.

Moved by Wm. J. Regan that a committee be appointed by the chairman to outline investigations which should be made regarding factors affecting official tests. Motion seconded and carried.

Annual election of officers.

The following officers were elected by unanimous ballot:

Chairman.....	H. P. Davis, Nebraska
Vice chairman.....	L. H. Fairchild, Indiana
Secretary.....	Roy T. Harris, Wisconsin

Adjournment followed.

R. B. BECKER,
Temporary Secretary.

ABSTRACT OF THE REPORT OF THE MEMBERSHIP COMMITTEE

J. A. Gamble, Maryland, Chairman

The first effort of your committee was to assemble, from available lists, the eligibles in the United States and Canadian Departments of Agriculture, the different state colleges and experiment stations, and other bodies so that we might know the total number from which we had to draw in carrying on a campaign, looking toward getting the leaders in dairy husbandry work in the field of teaching, extension and research into the American Dairy Science Association. At that time, articles 3 and 4 regarding membership read as follows:

MEMBERSHIP

Article 3. Membership shall be of two kinds, active and associate.

Article 4. The following are eligible for election as active members:

(1) Any person who is formally announced by an Agricultural College or Experiment Station or by the Dairy Division of the United States or Canadian Departments of Agriculture as an instructor, extension worker, investigator, or administrative officer connected with the dairy industry.

(2) Anyone filling a position of responsibility connected with the dairy industry of a professional character requiring technical scientific training.

Article 5. An active member ceasing active duties in connection with the dairy industry is transferred to associate membership. Any person interested in the dairy industry but not eligible under the qualifications prescribed for active membership may be nominated for associate membership. Associate members shall be entitled to all the privileges of the society except that of voting.

There were over 1200 such workers in the United States eligible under the above outline. This list disclosed that a large number of the leaders in the several fields were not as yet members of this body.

The following material, showing the scope of the Association, together with an application for membership was printed after the approval of the President and sent to all eligibles not appearing on the list of members furnished the committee by the Secretary of the Association.

THE AMERICAN DAIRY SCIENCE ASSOCIATION

The membership committee cordially invites you to be one of the new members who are joining our ranks this year.

The American Dairy Science Association is the professional organization of the dairy workers of this continent bearing to our profession somewhat the same relationship as the American Medical Association bears to the physician and the American Chemical Society to the chemist.

Ours is a working organization as well as a forum for discussion and an agent for the dissemination of scientific information. Its committees are constantly active in the improvement and standardization of technic, the improvement in dairy instruction, the stimulation of scientific research in all phases of the subject and improvement in methods of conduction of extension work.

The American Dairy Science Association is the most powerful single force in this continent for the advancement of the science of dairy husbandry. The Association holds an annual meeting usually in connection with the National Dairy Show, which furnishes an opportunity for all dairy husbandry workers of the country, whether their special interests lie in the field of teaching, research or extension, or handling and manufacture, to meet each other and to see the latest in dairy machinery and the best in dairy cattle.

Every two months we publish the *Journal of Dairy Science*, unique in its scope among the scientific publications of the English-speaking world. The *Journal of Dairy Science* is a medium for the publication of original communications, brief or extended, dealing with dairy husbandry problems. Its broad scope includes market milk handling, cheese making, ice cream manufacture, condensed milk, milk powder, butter making, nutrition, dairy cattle breeding problems, dairy chemistry, dairy bacteriology, dairy management, and dairy engineering.

The Association has at present time over 200 members. Before the end of the present year, it is hoped that our membership will be over five hundred members. The dues of the Association are \$5.00 a year, this including a year's subscription to the *Journal*. An application is attached to this letter which may be filled out and mailed in the enclosed envelope to J. A. Gamble, Chairman of Membership Committee, University of Maryland, College Park, Maryland.

MEMBERSHIP COMMITTEE OF THE AMERICAN DAIRY
SCIENCE ASSOCIATION

This was followed up by a circular, containing friendly comments on our *Journal of Dairy Science* prepared by Williams & Wilkins Company, Baltimore, Maryland, and setting forth the scope and the value of our magazine to those receiving the same. These comments were by the heads of the dairy departments in the different state colleges, by the officials of the breed associations, and by the different dairy companies and manufacturing supply houses.

The third step was the sending of a circular in the "Get a Member" campaign, to all men on the association rolls.

This circular made an urgent appeal to each member to get in touch with some fellow-worker eligible for membership and after acquainting him with the nature and scope of the work of the American Dairy Science Association, to extend to him a cordial invitation to membership.

This circular also stated the rules for eligibility as set forth in the constitution and urged members to take prompt action in order that all applications might be in before the annual meeting.

An "Application for Membership" blank was enclosed. The circular was followed up in ten days by a reminder urging members to send in the name of a new member so that a hundred per cent efficiency campaign might be reported at the St. Paul meeting.

At the St. Paul meeting, held in connection with the National Dairy Show, October 8th to 12th, 1921 the eligibility rules were changed to read as follows: thus doing away with the associate membership.

MEMBERSHIP

"Article 3. Membership shall be of one kind only; namely, Active.

"Article 4. The following are eligible for election:

"Any person who is formally announced by an agricultural college, or experiment station, or by the Dairy Division of the United States or Canadian Departments of Agriculture as an instructor, extension worker, investigator, or administrative officer connected with the dairy industry, or anyone filling a position of responsibility connected with the dairy industry and who has had a college or university training in technical science, or anyone filling a responsible position in the industry of a professional character requiring a technical knowledge of dairying of a high order."

On this basis, a list of the eligibles in the different sections was prepared and sent to the sectional members of the committee for the purpose of getting into the Association, the qualified men in the industry. The committee found that a great many men hesitated to come into the organization for the reason that they were unable to attend the annual meeting held in connection with the National Dairy Show. At this meeting, in St. Paul, permission was granted for the formation of sectional groups of the American Dairy Science Association. This will have a tendency to overcome this objection on the part of these men. Inasmuch as invitations have been extended to eligibles for active and associate membership under the old rules, the sectional leaders were instructed to accept applications under the old interpretation, but to make the new interpretation of eligibles their basis for additional work along this line.

The executive committee have ruled that the names of all eligibles shall be submitted to and have the approval of each member of the executive committee of the Association, before membership in the Association is offered to them. Under this plan, membership forms are to be prepared by the Association. These are to be furnished to the different sectional members of the committee who will send them to the heads of Dairy Husbandry Departments in the different Agricultural Colleges so that these heads may nominate for membership, any on their staffs or associated with them whom they deem eligible for nomination for active membership in the Association. These forms will be returned to the president of the Association through the chairman of the committee at College Park, Maryland. All these whom the executive committee deem desirable will receive a direct invitation to affiliate with us.

Results

As a result of the activities of the membership committee and many members of the Association, the committee is able to report that the applications of 110 were handled directly by the membership committee. This number does not include the applications sent direct to the president or secretary of the organization, nor does it include the back dues forwarded through the committee. The 110 men meant checks totaling \$550. It means also that the Association is now in a better position to serve its members at no increased cost.

The committee desires especially to thank Dr. Eckles, president of the Association, Professor Mortensen, the retiring president, and all other workers who have assisted in making the above results possible.

We wish to especially thank Mr. C. C. Thomas and the Williams & Wilkins Company who have furnished the printing for this campaign gratis. Also, the Dairy Husbandry Department of the University of Maryland, which furnished the material, did a large share of the work, and with the assistance of the Association, subsidized the membership drive.

H. F. JUDKINS,
W. H. E. R. REID,
THOMAS WRIGHT,
V. D. CHAPPELL,
J. A. GAMBLE,
Committee.

REPORT OF COMMITTEE ON DAIRY SHORT COURSE INSTRUCTION

J. R. Keithley, Minnesota, Chairman

Many of our colleges of agriculture offer instruction relative to the dairy industry. This instruction is, in most cases, divided into two classes, one of which is principally concerned with problems of milk production, the other with problems of handling, manufacturing and marketing of dairy products. This instruction is generally presented to three rather distinct classes or groups of students, whose needs requirements and fundamental training differ distinctly as follows: (a) *The collegiate student* obtains this instruction only as a part of his university education. (b) *The student in the secondary schools of agriculture* receives this instruction also only as a part of his more generalized practical agricultural education. (c) *The true short course student* in dairying devotes all of his time to and receives instruction only in dairying or some one specialized phase of it.

It was the understanding of the committee that only the *true short course instruction* was to be considered at this time. A study of the survey obtained by a questionnaire shows that these courses vary in their nature, and rightly so. This variation is due to local or sectional demands or needs. In some sections this demand is more urgent from the production field, in other sections it is more urgent from the manufacturing side. A summary of the questionnaire survey shows that the answers to the questions were as follows:

1. Does your college offer any short courses in Dairy Husbandry? If so, what courses are offered?

Twenty-two answered in the negative and twenty-six in the affirmative.

Those answering in the negative were: Alabama, Arizona, Arkansas, Colorado, Connecticut, Delaware, Florida, Louisiana, Maryland, Mississippi, Nevada, New Hampshire, New Jersey, New Mexico, North Carolina, North Dakota, Rhode Island, South Carolina, Texas, Virginia, West Virginia, Wyoming.

Those answering in the affirmative were: California, Georgia, Idaho, Illinois, Indiana, Iowa, Kansas, Kentucky, Maine, Massachusetts, Michigan, Minnesota, Missouri, Montana, Nebraska, New York, Ohio, Oklahoma, Oregon, Pennsylvania, South Dakota, Tennessee, Utah, Vermont, Washington, Wisconsin.

2. How many students were enrolled in each dairy short course offered by your college during the past year?

The number varied from 3 to 100 in each course and from 3 to 190 in each college or university. The total number of dairy short course students was 1149. The length of courses vary from one week to six months.

3. What are the aims of such courses? Are those aims being attained?

These aims vary somewhat and are as follows:

a. Through a knowledge of theory and practice to produce more capable helpers for the ice cream, butter, cheese and milk plant operators and owners.

b. To train home demonstration agents to instruct farm women in best methods of handling dairy products and in the care and management of dairy cows.

c. To present to farmers some of the most important problems of the present day and discuss problems which they themselves bring up.

d. To acquaint young men with livestock so they may prove more useful men on farms and will work toward positions as herdmen, managers, and eventually owners.

e. To teach the elements of dairying relative to breed, selection, breeding, feeding, care and management, as well as manufacturing of dairy products.

f. To offer instruction to men and women who wish to take up general farming.

g. To prepare specialists in dairy manufactures so as to be capable of taking charge of small creameries, milk plants, cheese factories, or of ice cream factories.

h. To prepare men to act as supervisors for A. R. O. and cow testing association work.

i. To instruct the producers concerning the essentials in producing and delivering a sanitary milk or cream to the dealers.

4. Will you please offer suggestions as to ways in which you consider it possible to make improvements in this type of dairy courses?

Suggested improvements are as follows:

a. Divide Short Courses on basis of (1) Care and feeding of cows, (2) handling and manufacture of dairy products.

b. Operate a commercial plant using up-to-date equipment. Admit only experienced man (one year) and have special instructors.

c. Present only material having a direct bearing and make this concise and to the point. Have first-class practical men assist in instruction giving all laboratory work possible. Limit the theoretical work.

d. Lengthen the courses. In production work include judging. In manufacturing courses cover fewer subjects.

e. Have scholastic entrance requirements as well as experience requirement. Lecture method is best for experienced men, but textbook and recitation method is best in the longer courses for the less experienced men.

f. Instead of general course in dairy manufactures, specialized courses in each of the following should be given: butter, cheese, ice cream, and market milk.

5. What principal difficulties have been encountered by your institution in giving dairy short courses instruction?

The principal difficulties mentioned were as follows:

a. In many states demand is not sufficient to warrant dairy short courses.

b. Lack of facilities, in men, money, and equipment.

c. Creates too heavy teaching load for regular college staff because short course work is added to the regular college and school duties.

d. Presenting material to experienced and inexperienced men in same class in such a way as to satisfy both.

e. Selecting suitable seasons of year for representative work.

f. Lack of good teachers.

6. Will you please send descriptive bulletins, circulars, catalogs, or other statements relative to short courses?

Twenty institutions sent descriptive bulletin or materials.

7. Do you follow the lecture method of instruction for short course students?

The method of instruction, in most cases, was lecture and laboratory supplemented by assigned readings, recitation and discussion.

8. To what extent do you use a textbook? What textbooks do you use for short course students?

Answers indicated that textbooks used varied with the nature of the short course offered.

9. To what extent is the instruction given by the regular college staff and special instructors?

Answers indicated that short course instruction is given by the regular college staff. The following states have additional special instructors: California, Illinois, Iowa, Minnesota, New York, Ohio, Oregon, Pennsylvania, Vermont, Washington and Wisconsin.

10. How are the instructors paid? In answer:

a. The regular staff members are not paid additional for short course work.

b. The special instructors receive expenses, and in some cases, \$7.00 to \$15.00 daily.

1. To what extent are products handled or manufactured in these courses?

Answers indicated that products are made only on a small scale, to illustrate principles, by colleges not having commercial plant. Those having a commercial plant make somewhat large quantities but for the same purposes. Some churn three or four times daily, and make up cheese from 1200 to 1500 pounds milk daily.

Ice cream is made in from 50- to 200-gallon lots per laboratory exercise.

12. Will you please supply us with lecture and laboratory outlines followed by students in these courses?

In response, lecture and laboratory outlines were submitted by Massachusetts, New York and Pennsylvania. Others have no definite outlines available but vary their programs to meet desires of students enrolled.

The Committee would recommend:

1. Adoption of more uniform, standardized short course instruction among the colleges and universities where desirable and possible.

2. Such uniformity and standardization would be aided through definite lecture and laboratory outlines being made available.

3. A wider use of available textbooks to supplement lectures and laboratory work would be desirable.

4. Sufficient laboratory work be required to fix clearly in the student's mind the principles involved in the work under consideration.

H. W. GREGORY, Indiana,

P. M. BRANDT, Oregon,

J. R. KEITHLEY, Minnesota,

Committee.

REPORT OF COMMITTEE ON LEGAL STANDARDS AND SCORE CARDS FOR DAIRY PRODUCTS

J. H. Frandsen, Nebraska, Chairman

Your Committee on Score Cards for Dairy Products has had considerable correspondence and sub-committees have held a number of meetings in which much attention has been given to new score cards. This year most of the time has been given to consideration of score cards for sweetened condensed milk, evaporated milk and ice cream.

Regarding sweetened condensed milk, your committee desires to recommend a tentative score card consisting of the following items:

SCORE CARD FOR SWEETENED CONDENSED MILK

<i>Properties</i>	<i>Perfect score per cent</i>
Flavor and odor.....	30
Body and texture.....	25
Color.....	5
Fat content.....	10
Milk solids.....	10
Bacteria.....	10
Sugar.....	10
Adulterants and preservatives (must be absent)	—
Total score.....	100

Suggestions for use of score card

Flavor and odor. Perfect; must be fresh, sweet and free from all flavors. Deduct one to ten points each if metallic, rancid, stale, cheesy. Deduct one to thirty points if sour, yeasty or otherwise fermented.

Body and texture

Perfect: Must be viscous, smooth and free from lumps of curd, sugar sediment and foreign impurities. Deduct one to five points if rough and sandy, from one to five points if sugar sediment in bottom, from one to five points if fat separation, one to five points if white and yellow buttons, 15 to 25 points if lumps of curd.

Color

Perfect: Rich cream to yellow. Deduct one to five points if brown.

Fat content

Perfect: Must contain not less than 10 per cent milk fat. Deduct one point for each half per cent less than 10 per cent. If below 8 per cent deduct ten points. Deduct ten points if below 8 per cent present Federal Standard.

Total milk solids

Perfect: Must contain not less than 32 per cent. Deduct one point for each per cent or fraction thereof below 32 per cent. If below 28 per cent deduct ten points.

Sugar

Perfect: The concentration shall be from 60 to 62 per cent. Deduct two points for each per cent concentration below 60 or above 62 per cent. The concentration shall be determined by *dividing* per cent of sugar by the sum of per cent of sugar and water.

Bacteria

Make reduction for excessive number of bacteria. Importance of bacterial counts has not as yet been sufficiently considered by the committee to warrant definite recommendations.

SCORE CARD FOR EVAPORATED MILK

Regarding evaporated milk, the following score card is tentatively suggested.

<i>Properties</i>	<i>Perfect score per cent</i>
Flavor and odor.....	40
Body and texture.....	35
Color.....	5
Fat content.....	10
Total solids.....	10
Adulterants and preservatives must be absent	—
Total score.....	100

Flavor and odor. Perfect: Must be fresh, sweet and free from off flavors. Deduct 1 to 10 points if metallic, rancid and stale. Deduct 40 points if sour, bitter, putrid, gassy or otherwise fermented.

Body and texture. Perfect: Must be creamy, of uniform emulsion, smooth. Deduct 1 to 10 points each for curdy milk, separated or churned milk.

Color. Perfect: Must be creamy. Deduct 1 to 3 points if brown.

Fat content. Perfect: Must contain not less than 9 per cent milk fat. Deduct one point for each one-half per cent less than 9 per cent. Deduct 10 points if less than 7.8 per cent the present Federal Standard.

Total solids. Perfect: Must contain not less than 28 per cent solids. Deduct 1 point for each 1 per cent or fraction thereof, less than 28 per cent. Deduct 10 points if below 25.5 per cent, present federal standard.

Adulterants and preservatives. Perfect: Must be free from all adulterants or preservatives. If it contains animal or vegetable fats, or other ingredients foreign to the composition of normal milk, or any preservatives deduct 100 per cent.

MILK POWDER

The committee has under consideration a score card for milk powder but do not feel justified in making a definite recommendation at this time.

ICE CREAM

Regarding ice cream score card, your committee recommends that the following score card be tentatively recommended for the coming year and that all interested users furnish the committee with their suggestions regarding same so that during the year they may have more available data on which to base further recommendations.

SCORE CARD FOR ICE CREAM

Flavor.....	40
Body and texture.....	25
Fat and solids.....	10
Bacteria.....	20
Package.....	5
Total.....	<u>100</u>

MILK SCORE CARD

Your committee, in the absence of data supporting a change, recommend the adoption of the milk score card tentatively used by the Ameri-

can Dairy Science Association this year and which carries the following items:

Bacteria.....	35
Flavor and odor.....	25
Fats.....	10
Solids-not-fat.....	10
Sediment.....	10
Acidity.....	5
Bottle and cap.....	5
	<hr/>
Total.....	100

The above report has the approval of all members of the score card committee present at meeting.

O. F. HUNZIKER,
L. A. ROGERS,
J. A. GAMBLE,
W. P. B. LOCKWOOD,
J. H. FRANDSEN,
Committee.

As chairman of the committee, I desire at this time to acknowledge my appreciation for the fine work done by Professors O. F. Hunziker and L. A. Rogers, who constituted the sub-committee doing most of the work on condensed milk score card, and to Professors J. A. Gamble and W. P. B. Lockwood for their work on milk score card, and of all the members of the Committee who assisted in the preparation of this report.

J. H. FRANDSEN,
Chairman.

REPORT OF COMMITTEE ON STUDENTS' NATIONAL CONTEST IN JUDGING DAIRY PRODUCTS

A. W. Rudnick, Iowa, Chairman

The Students' National Contest in Judging Dairy Products shall be a contest among agricultural students—individuals and teams—in judging the quality and market grade of butter, cheese and milk.

Ten samples of each product shall be scored, placed and criticized. This contest shall be held annually at the National Dairy Show and shall be under the direct supervision of the Dairy Division, United States Department of Agriculture, a member of which division shall be the superintendent of the contest.

Any student of a land-grant agricultural college, or of a secondary school under direct supervision of a land-grant state agricultural college, who is regularly matriculated in a course of at least two years in agriculture or dairying and has taken not less than twelve weeks undergraduate work during the calendar year in which the show is held, who has never taken part in any contest for judging dairy products of a national or international character, who has never acted as an official judge of dairy products, and who has not taught the manufacture of, or the judging of dairy products, may enter as a member of a team. Three men from any one college or school shall constitute a team.

RULES GOVERNING CONTEST

1. All entries must be received by Mr. W. E. Skinner, General Manager of the National Dairy Show, not later than one week previous to the date of contest.

Regular entry forms will be mailed in due time to the professor of dairying, or dairy manufacturing, in each state agricultural college.

2. An entry fee of two dollars (\$2.00) is charged each contestant, and this must be forwarded with the entry form. The money received from this source is used in helping defray expenses of the contest.

3. Each institution eligible to participate in this contest may enter a team, consisting of three eligible students of that institution.

4. Each contestant shall report to the superintendent of the contest at such time and place as may be announced. He will then be assigned a number and given such instructions as the superintendent may deem necessary.

5. The contestants shall be divided into three groups, each consisting of one member of each team.

6. No contestant shall wear any uniform, college colors, college hat or college pin which may in any way reveal his identity or the identity of the college which he represents.

7. No contestant shall be allowed to take any note-book or writing paper into the contest, except such cards as are provided by the superintendent of the contest.

8. While the contest is in progress, there shall be no communication between the contestants or between contestants and anyone else, except as directed by the superintendent or his representative, and then only in the presence of the superintendent or his representative.

9. Reporters, officials and others, except the contestants, the superintendent and his assistants, shall be excluded from the place of scoring while the contest is in progress.

10. Any contestant violating any rule will be debarred from the contest. If a member of any team is debarred because of violation of rules, that team will be debarred from the team contest, although the remaining members may compete for the individual prizes.

11. Each contestant, on entering the place of contest will be given score cards on which to record his scores and criticisms. At the expiration of the allotted time all score cards shall be returned to the superintendent. The contestant shall place on each card the number assigned to him.

12. Dairy products will be scored on the following basis:

<i>Butter</i>		<i>Cheese</i>		<i>Market milk</i>	
Flavor.....	45	Flavor.....	45	Flavor and odor....	25
Body.....	25	Texture.....	30	Sediment.....	10
Color.....	15	Color.....	15	Bottle and cap.....	5
Salt.....	10	Make-up.....	10		
Package.....	5				
<hr/>		<hr/>		<hr/>	
Total.....	100	Total.....	100	Total.....	40

The butter, as far as practicable, shall be scored and criticised in accordance with the system outlined in Service and Regulatory Announcement No. 51 of the Bureau of Markets, United States Department of Agriculture.

13. Each contestant shall score and criticise ten tubs of butter, ten cheese, and ten samples of milk, the time allowance being one hour for each product.

14. No member of the teaching, extension or experimental staff of a college entering a team, may act as an official or be in any way connected with the Dairy Products exhibit of the National Dairy Show.

15. Each contestant shall place the samples in order according to their total scores. Placing the number of the highest sample first, the second highest second, etc.

16. The superintendent shall set out not more than five representative samples of each product on the afternoon preceding the contest. The time to be specified by the superintendent. Not more than one such exhibit shall be prepared and all contestants shall be notified of the time at least ten days in advance.

SUPERINTENDENT OF CONTEST

It shall be the duty of the superintendent of the contest to see that all rules and regulations governing the contest are duly carried out, and shall see that the contest is conducted with fairness and justice to all concerned.

He shall have a sufficient number of clerks and assistants to help him in conducting the contest.

He shall direct the contestants as to the products to judge, time to commence work and time to stop.

After instructing the contestants in a body regarding the contest, the form in which to prepare the scores and criticisms, etc., he shall say nothing to any contestant as to method for the contestant to follow, in scoring.

He shall have charge of all records and shall have all ratings tabulated and totaled, and shall deliver the results of the contest to the general manager of the National Dairy Show.

He shall decide all questions which may arise in connection with the interpretation of the rules governing the contest, and may make additional rulings to facilitate the working of the general rules.

CLERKS AND ASSISTANTS

One assistant shall have charge of each group of contestants and shall see that each contestant in his group remains in his presence the entire time the contest is in progress; except in cases of emergency, and then as directed by the superintendent.

Clerks shall also be provided for the judging committee.

PRODUCTS AND MARKING

The products shall be provided by the National Dairy Association.

A member of the United States Department of Agriculture shall select ten tubs of butter, ten cheeses, and ten samples of market milk which in his judgement will make a representative scoring contest for each product.

The packages shall be numbered consecutively and the numbers be marked plainly on the packages.

JUDGES

The official scores and criticisms shall be placed on these products by a committee consisting of a commercial judge and judge from the United States Department of Agriculture, for butter, and a commercial and United States Department of Agriculture judge for cheese, and two United States Department of Agriculture, or other judges for milk. The products shall be judged on the following basis:

<i>Butter</i>		<i>Cheese</i>		<i>Market milk</i>	
Flavor.....	45	Flavor.....	45	Flavor and odor....	25
Body.....	25	Texture.....	30	Sediment.....	10
Color.....	15	Color.....	15	Bottle and cap.....	5
Salt.....	10	Make-up.....	10		
Package.....	5				
<hr/>		<hr/>		<hr/>	
Total.....	100	Total.....	100	Total.....	40

The butter, as far as practicable, shall be scored and criticised in accordance with the system outlined in Service and Regulatory announcement No. 51 of the Bureau of Markets, United States Department of Agriculture.

It shall be the duty of the judge to score each sample of the product which he is judging and write his score and criticisms on cards that will be provided. Score cards must be numbered to correspond with numbers on the packages (tubs of butter, cheese, or bottles of milk) and be signed by the judge. He may keep a memorandum of his scores and criticisms.

The score cards will be collected by clerks and the scores averaged. The average of the scores of the judges will be the "official score" for each sample respectively. Reasons of the judges will be the "official reasons."

If the score on any product varies by more than one point on flavor or more than one half point on any other item or more than one point in total, and in case that the judges do not come within these limits, a new sample shall be submitted.

The criticisms shall agree in all particulars. Each judge shall satisfy himself that defect or other fault called attention to by the other judge is apparent to himself, or such defect should not be noted, or new sample called for in case of disagreement.

There shall be no tie scores. If samples cannot be scored off new samples shall be submitted.

The judge of market milk shall prepare regulation sediment discs, which shall be displayed in a petri dish or other suitable manner, be numbered and next to the milk samples being judged.

TO INSURE A UNIFORM STANDARD

No samples shall be open in the room at the time of scoring excepting those being judged. Uniformity shall be worked out as prescribed heretofore.

GRADING

The work of the contestants shall be graded on the following. The scores, criticisms and placing shall be graded separately but the grades shall be added to form the total for each product which shall be expressed negatively. The grades shall be established by a grading committee consisting of the superintendent of the contest and such assistants as he may select. The judges shall grade all criticisms and shall take the scores and statements into consideration when making the grade on criticisms.

BUTTER

Score

Flavor. The contestant's score on flavor will be given a grade expressed by the difference between his score and the official score.

Body. Scored as for flavor.

Color. Scored as for flavor.

Salt. Scored as for flavor.

Package. Scored as for flavor.

Placing

The score for placing shall be determined by the sum of the differences in the position of the contestant's samples and the official placing.

Total scores only will be taken into consideration. Example: If the contestant places the highest scoring example 4th, he would lose 3 points; the second highest sample 7th, he would lose 5 points and so on for all ten samples. The sum of the differences shall be the score on placing.

Criticism

The contestant shall not be graded off more than 5 on any one sample for criticism.

DETERMINING THE GRADE

A contestant's grade shall be found by adding the differences on scoring, placing and criticism. The contestant having the lowest score shall be first. A team's grade is the sum of the grades of its members. The teams are placed according to their grades, the team having the lowest score wins first place.

When two or more contestants tie or teams have the same grade, the awarding of the medals shall be decided by determining which has the highest score on flavor, then on body, then on color, then on salt, then on package. If no prize is involved, the placing shall be given as a tie.

CHEESE

Score

Flavor. The contestant's score on flavor will be given a grade expressed by the difference between his score and the official score.

Body. Scored as for flavor.

Color. Scored as for flavor.

Salt. Scored as for flavor.

Make-up. Scored as for flavor.

Placing

The score for placing shall be determined by the sum of the differences in the position of the contestants' samples and the official placing. Total scores only will be taken into consideration. Example: If the contestant places the highest scoring sample 4th he would lose 3 points; the second highest sample 7th, he would lose 5 points and so on for all ten samples. The sum of the differences shall be the score in placing.

Criticism

“Criticism” in cheese judging is graded in the same way as that described under butter judging.

DETERMINING THE GRADE

Same as for butter.

MILK

Score

Flavor and odor. The contestant's score on flavor will be given a grade expressed by the difference between his score and the official score.

Sediment. Scored as for flavor.

Bottle and cap. Scored as for flavor.

Criticism

Criticism in milk judging is graded in the same way as that described under butter judging.

DETERMINING THE GRADES AND PLACINGS

Same as for butter.

Placing of contestants on all products. The contestant whose sum of grades on the three products is lowest wins first place in judging all products.

Placing of teams on all products. The team whose sum of grades on the three products is lowest wins first place in judging all products.

Ties in placing contestants and teams on all products shall be broken in the manner described under “butter,” if possible.

REPORT OF COMMITTEE ON OFFICIAL METHODS FOR TESTING MILK AND CREAM FOR BUTTERFAT

O. F. Hunziker, Chicago, Chairman

Your committee has given careful consideration to the subject of official methods of testing milk and cream for butterfat and beg to submit the following report:

1. We recommend that no changes nor modifications be made in the official and standard methods of testing milk and cream for butterfat and in the standard specifications for Babcock testing glassware, as adopted by this Association at its annual meeting in 1916 and as modified and approved by this Association at its annual meeting in 1920.

2. Your committee desires to call attention, however, to the regretful fact that there are many instances in the operation of the Babcock test where such factors as proper speed and proper temperature of the Babcock centrifuge are seriously neglected and frequently entirely ignored, thus jeopardizing the accuracy of the results.

Recent findings by H. B. Siegmund and R. Sewell Craig, published in the *Journal of Dairy Science*, volume IV, number 1, January, 1921, demonstrate anew the importance of proper speed of the tester. These findings indicate that not only is a certain minimum speed necessary to separate and assemble in the neck of the bottle all the butterfat, but the speed of the tester must be sufficient to remove from the fat column excessive admixture of water and acid.

Because of this tendency on the part of the operator to neglect the attention to proper speed and temperature of the centrifuge, your committee respectfully urges that those engaged in the instruction of testing milk and cream place special emphasis on the importance of proper speed and proper temperature of the tester, and insist that Babcock centrifuges be equipped with steam gauge (in case of steam driven machines) automatic speed indicator, adequate heating device, and thermometer.

3. At the last annual meeting your committee was instructed to give greater publicity to the official methods adopted by this Association. Inasmuch as the committee's report approved at last year's meeting and containing modifications of the official methods previously adopted, failed to appear in the columns of the *Dairy Science Journal*, your Committee recommends that a complete copy of the specifications for

standard Babcock glassware and of the official methods for testing milk and cream for butter fat, as adopted in 1916, and embracing the modifications adopted in 1920, be published in an early issue of the *Dairy Science Journal*, and that the 25 copies which are furnished gratis with each article, by the printers, be placed at the disposition of the Chairman of your Committee.

4. Your committee has also had under investigation methods for testing buttermilk and skimmilk for butterfat with the view of devising a method that we could recommend to this Association for adoption as a standard or official method.

As the result of this investigation and of the consideration of diverse methods of testing these by-products, your committee begs to present to this Association a method devised by Prof. Mitchell of the American Association of Creamery Butter Manufacturers. We feel that this method is a vast improvement over the ordinary Babcock Test. Its results appear to be uniform and to check closely with those of the Roesse Gottlieb method. The directions given by Prof. Mitchell for the above, are as follows:

ACID-ALCOHOL TEST

Directions for testing buttermilk

Chemicals. Commercial sulphuric acid, normal butyl alcohol.

1. Add the chemicals and buttermilk to the test bottle in the following amounts and the order indicated.

- (a) 2 cc. of N-butyl alcohol.
- (b) 9 cc. of buttermilk.
- (c) 7 cc. of commercial sulphuric acid.

Vary amount of acid to suit its strength. The right amount is being used when the fat column is golden yellow to light amber in color.

- 2. Mix contents of bottle thoroughly.
- 3. Centrifuge for six minutes.
- 4. Add hot water (soft or distilled) to fill bottle to bottom of neck and whirl two minutes.
- 5. Add balance of water to float fat into neck and again whirl two minutes.
- 6. Read at temperature of 135°-140°F. Double the reading to obtain per cent of fat.

7. In cleaning test bottle—especially if there be any deposit—first add a small amount of luke-warm water and to this add sulphuric acid. Always add the water first and then the acid—never the reverse. Rinse the bottle well with this mixture and then rinse with hot water.

This test gives results corresponding to those of chemical analysis.

A test bottle, with a scale reading up to 0.50 per cent for 18 grams, should be used.

We understand that butyl alcohol is to be preferred to amyl alcohol because it is comparatively easy to obtain industrial butyl alcohol free from impurities, while such is not always the case with amyl alcohol.

5. Because of the highly satisfactory results obtained by the use of the acid-alcohol test, your committee recommends that this new method be given a thorough trial, during the ensuing year, by those members of the Association who are in position to test buttermilk, and that if possible their results be reported to the Committee. While the acid-alcohol test has been worked out largely on buttermilk only, there is every indication that it will prove equally satisfactory with skimmilk. We, therefore, further recommend that this trial include skimmilk as well as buttermilk.

6. Your Committee begs to also call attention to the modified Babcock test recommended by Profs. T. J. McInerney and H. C. Troy, as published in Cornell Bulletin No. 401, January, 1920. McInerney and Troy recommend the following procedure:

1. At least 25 cc. of sulphuric acid should be used. If the size of the bottle permits, as much as 28 cc. may be used to advantage.

2. The temperature of the testing machine should be at least 180°F.

3. A centrifuge having a disk 15 inches in diameter, and strong enough to be run at a minimum speed of 1800 revolutions a minute without danger of breaking, is recommended. The diameter of the disk is determined by measuring the distance between the bases of the opposite cups when they are in a horizontal position.

4. The milk should be centrifuged for ten-, two-, and one-minute periods, instead of for five-, two-, and one-minute periods.

The above directions are given for skimmilk only.

F. W. BOUSKA,
L. A. ROGERS,
R. H. SHAW,
H. C. TROY,
O. F. HUNZIKER,
Committee.

SPECIFICATIONS AND DIRECTIONS FOR TESTING MILK AND CREAM
FOR BUTTERFAT¹

O. F. Hunziker

I. APPARATUS AND CHEMICALS

Milk test bottle. 8 per cent 18 gram milk test bottle, graduated to 0.1 per cent. Graduation. The total per cent graduation shall be 8. The graduated portion of the neck shall have a length of not less than 63.5 mm. (2½ inches). The graduation shall represent whole per cent, five-tenths per cent and tenths per cent. The tenths per cent graduations shall not be less than 3 mm. in length; the five-tenths per cent graduations shall be 1 mm. longer than the tenths per cent graduations, projecting 1 mm. to the left; the whole per cent graduation shall extend at least one-half way around the neck to the right and projecting 2 mm. to the left of the tenths per cent graduations. Each per cent graduation shall be numbered, the number being placed on the left of the scale.

The maximum error in the total graduation or in any part thereof shall not exceed the volume of the smallest unit of the graduation.

Neck. The neck shall be cylindrical and of uniform internal diameter throughout. The cylindrical part of the neck shall extend at least 5 mm. below the lowest and above the highest graduation mark. The top of the neck shall be flared to a diameter of not less than 10 mm.

Bulb. The capacity of the bulb up to the junction of the neck shall not be less than 45 cc. The shape of the bulb may be either cylindrical or conical with the smallest diameter at the bottom. If cylindrical, the outside diameter shall be between 34 and 36 mm.; if conical, the outside diameter of the base shall be between 31 and 33 mm., and the maximum diameter between 35 and 37 mm.

The charge of the bottle shall be 18 grams.

The total height of the bottle shall be between 150 and 165 mm. (5½ and 6½ inches).

Cream test bottle 1. 50 per cent 9 gram short-neck cream test bottle, graduated to 0.5 per cent. Graduation—The total per cent graduation shall be 50. The graduated portion of the neck shall have a length of not less than 63.5 mm. (2½ inches). The graduation shall represent 5 per cent, 1 per cent, and 0.5 per cent. The 5 per cent graduations

¹ Published by recommendation of Committee on Official Methods for Testing Milk and Cream for Butterfat, and approved by American Dairy Science Association.

shall extend at least half-way around the neck (to the right). The 0.5 per cent graduations shall be at least 3 mm. in length, and the 1 per cent graduations shall have a length intermediate between the 5 per cent and the 0.5 per cent graduations. Each 5 per cent graduation shall be numbered, the number being placed on the left of the scale.

The maximum error in the total graduation or in any part thereof shall not exceed the volume of the smallest unit of the graduation.

Neck. The neck shall be cylindrical and of uniform internal diameter throughout. The cylindrical part of the neck shall extend at least 5 mm. below the lowest and above the highest graduation mark. The top of the neck shall be flared to a diameter of not less than 10 mm.

Bulb. The capacity of the bulb up to the junction of the neck shall not be less than 45 cc. The shape of the bulb may be either cylindrical or conical with the smallest diameter at the bottom. If cylindrical, the outside diameter shall be between 34 and 36 mm.; if conical, the outside diameter of the base shall be between 31 and 33 mm. and the maximum diameter between 35 and 37 mm.

The charge of the bottle shall be 9 grams. All bottles shall bear on top of the neck above the graduations, in plainly legible characters, a mark defining the weight of the charge to be used (9 grams).

The total height of the bottle shall be between 150 and 165 mm. ($5\frac{7}{8}$ and $6\frac{1}{2}$ inches), same as standard milk test bottles.

Cream test bottle 2. 50 per cent 9 gram long-neck cream test bottle, graduated to 0.5 per cent. The same specifications in every detail as specified for the 50 per cent 9 gram short-neck bottle shall apply for the long-neck bottle with the exception, however, that the total height of this bottle shall be between 210 and 235 mm. ($8\frac{1}{4}$ and 9 inches), that the total length of the graduation shall be not less than 120 mm., and that the maximum error in the total graduation or in any part thereof shall not exceed 50 per cent of the volume of the smallest unit of the graduation.

Cream test bottle 3. 50 per cent 18 gram long-neck cream test bottle graduated to 0.5 per cent. The same specifications in every detail as specified for the 50 per cent 9 gram long-neck shall also apply for the 18 gram long-neck bottle, except that the charge of the bottle shall be 18 grams. All bottles shall bear on top of the neck above the graduation, in plainly legible characters, a mark defining the weight of the charge to be used (18 grams).

"Pipette, capacity 17.6 cc. of water at 20°C. Total length of pipette not more than 330 mm. ($13\frac{1}{2}$ inches). Outside diameter of suction tube

6 to 8 mm. Length of suction tube 130 mm. Outside diameter of delivery tube 4.5 to 5.5 mm. Length of delivery tube 100 to 120 mm. Distance of graduation mark above bulb 15 to 45 mm. Nozzle straight. To discharge when filled with water in 5 to 8 seconds. The maximum error shall not exceed 0.05 cc. In the operation of the test the last drop of milk should be blown out of the pipette into the test bottle."

Acid measure, capacity 17.5 cc.

Cream testing scales. Sensibility reciprocal of 30 mgm., i.e., the addition of 30 mgm. to the scales, when loaded to capacity, shall cause a deflection of the pointer of at least one division on the graduation.

Weights, 9 gram weights for 9 gram test bottles and 18 gram weights for 18 gram cream test bottles, preferably stamped correct by the United States or State Bureau of Standards.

Tester. Standard Babcock test centrifuge and speed indicator.

Dividers for measuring fat column.

Water bath for cream samples, with proper arrangement for regulating and recording temperature of samples.

Water bath for test bottles, of sufficient size and with necessary equipment to insure proper control of temperature. The following dimensions for a twenty-four bottle water bath are recommended: Metal box, 14 inches long, 11 inches wide and $8\frac{1}{2}$ inches deep and equipped with a bottle basket $9\frac{1}{2}$ inches long and $6\frac{1}{2}$ inches wide, capacity twenty-four bottles, a steam and water inlet, a drain a thermometer holder with thermometer.

Chemicals. Commercial sulphuric acid, specific gravity 1.82 to 1.83; glymol, or white mineral oil, high grade.

II. MANIPULATION OF TEST

A. Milk test

Milk samples. Single samples are preferred to composite samples. If composite samples are used they should be kept in clean jars sealed air-tight, and containing a sufficient amount of preservative. Corrosive sublimate, potassium bichromate and formaldehyde may be used as satisfactory preservatives. For the keeping of composite samples a cool location should be chosen. They should be the product of not over one week and should be tested as soon as possible.

If transported by mail, express or otherwise the sample bottle should be completely full and tightly stoppered and the samples should be preserved as above directed.

Immediately before testing the sample is thoroughly mixed until it is homogeneous. If lumps of cream, butter or ice do not completely disappear, heat to 100° to 120°F., mix thoroughly and pipette at once. Avoid incorporation of air bubbles while mixing the sample. Curdy and churned samples are not dependable.

Testing. Measure 18 grams of milk from properly mixed sample into standard milk test bottle, by using 17.6 cc. standard pipette; add 17.5 cc. of standard commercial sulphuric acid, and shake until all curd has disappeared, and then continue the shaking for a few moments longer. Milk and acid before mixing should have a temperature of 50° to 70°F.

Whirl in Babcock centrifuge for five, two and one minutes, respectively, filling the bottle with hot water (temperature 140°F. or above) to the bottom of the neck after the first whirling and to near top graduation after the second whirling. The proper speed of the centrifuge is 800 revolutions for an 18-inch diameter wheel and 1000 revolutions per minute for a 12-inch diameter wheel.

Set the test bottles into water bath and read after a temperature of 135°F. to 140°F. has been maintained for not less than 3 minutes. Read test by measuring fat column from bottom of lower meniscus to top of upper meniscus. Use dividers for reading.

B. Cream test

Cream samples. Cream samples should be tested as soon as possible and not later than three days after they are taken. Composite samples representing portions of consecutive deliveries of the same patron are unreliable. Samples should at all times be kept in nonabsorptive containers, sealed air-tight and held in the cold.

Immediately before testing mix the sample until it pours readily and a uniform emulsion is secured. If in good condition shake, pour or stir until properly mixed. If very thick, warm to 85°F. and then mix. In case of lumps of butter heat the sample to 100°F. to 120°F. by setting in water bath, mix thoroughly and weigh out at once. For commercial work on a large scale it is advisable to temper all samples to 100° to 120°F. in a water bath previous to mixing. Great care should be exercised to avoid overheating the sample, causing the cream to "oil off." This precaution is especially necessary with thin cream.

Testing. Weigh 9 grams or 18 grams, respectively, of the properly mixed sample into a standard cream test bottle on standard cream testing scales which are in proper working condition, set level and are protected from drafts.

Method I. Add standard commercial sulphuric acid until the mixture of acid and cream, immediately after shaking, resembles in color, coffee with cream in it. Usually about 8 to 12 cc. of acid is required in the case of the 9 gram or 14 to 17 cc. of acid in the case of the 18 gram bottle, the amount needed depending on the temperature of acid and cream and on the richness of the cream.

Whirl in Standard Babcock centrifuge at proper speed, five, two and one minutes, respectively, filling the bottles with hot soft water, temperature 140 F. or above, to the bottom of the neck after the first whirling and to near the top graduation after the second whirling.

a. Alternate Method. Add 9 cc. of water after the cream has been weighed into the test bottle and before the acid is added, then add 17.5 cc. acid and proceed as in previous method. This method is applicable with the 9 gram bottle only.

b. Alternate Method. Add 8 to 12 cc. of acid in the case of the 9 gram bottle or 14 to 17 cc. of acid in the case of the 18 gram bottle, or add acid until the mixture of cream and acid, after shaking, has a chocolate brown color. After the cream and acid have been thoroughly mixed and all lumps have completely disappeared, add a few cubic centimeters (not less than 5 cc.) of hot soft water, whirl five minutes, add hot soft water to near top of scale and whirl one minute.

The proper speed of the centrifuge is 800 revolutions per minute for an 18 inch diameter wheel and 1000 revolutions per minute for a 12 inch diameter wheel.

Set the test bottles into water bath and read after a temperature of 135°F. to 140°F. has been maintained for not less than three minutes, add a few drops of glymol and read at once, preferably using dividers. Experienced testers are able to secure correct readings without glymol by reading to the bottom of the upper meniscus but the use of glymol is urged.

C. Defective tests

The fat column of the finished test should be clear, translucent and should have a golden yellow to amber color. All tests which are milky, or foggy, or showing the presence of curd or charred matter in or below the fat column, or of which the reading is indistinct or uncertain, should be rejected. Duplicate tests are essential in all work where special accuracy of results is required, such as in official testing and experimental investigations.

REPORT OF COMMITTEE ON RELATION TO BREED ASSOCIATIONS

E. G. Woodward, Washington, Chairman

1. REPORT OF SUB-COMMITTEE ON ADMINISTRATION OF OFFICIAL TESTING WORK WITHIN THE VARIOUS STATES

A. Office records. These consist of (a) duplicate copies of all test reports to be filed at College under name of owner and name or H. B. number of cow. (b) Card or book record showing supervisor assigned, date of test and its length, cows reported, reports sent to club, and charge against breeders. (c) Markers or lists showing delinquent breeders or records needing special attention. (d) Ledger accounts with breeders and cattle clubs, cost sheets, vouchers, etc., appropriate to accounting system in use at the respective institutions. (e) Statistical data forms.

Recommendations. It is hardly necessary to say that each institution ought certainly to maintain a complete file of original test sheets. The clubs rightly expect this of us. Our system of office checking should be designed simply and should be so direct that the records will not be delayed in transmission. We advise that the committee take up with the clubs the question of forms to be furnished for billing check tests, etc., with a view to uniformity. It would also be worth while if some of us should be designated to receive samples of new forms or improvements devised by our members and to circulate suggestions among others. It has been suggested that uniform blanks be designed for use in the various States which could be printed in quantity and distributed from some central point. There would be some difficulty in financing such an arrangement. A committee could be appointed, perfect plans and get tentative bids if a sufficient number of States are interested.

B. Training supervisors. Results from special courses are reported disappointing in most cases. The number secured by this means is so small as to hardly pay for the time spent in giving the instruction while the quality is no improvement. The committee believes that better results are secured at no greater cost through personal coaching of candidates before first assignment, where they have taken their training in some college or secondary school course. Candidates may better be selected on basis of moral character, reputation in home community,

intelligence, experience, and general appearance and bearing, than to depend too much on training alone.

C. District organization. Indiana, Illinois and Wisconsin have attempted to cooperate in maintaining a supply of well-trained supervisors while avoiding the embarrassment of a surplus. Under such circumstances as have existed the past four years, the plan has worked well. At our present stage of development, we found it hardly practicable to include more than two states in such district. We worked informally. Our experience indicates that districts should be laid out in squares or with greatest length north and south, so that busy seasons in different sections will fall at different times. All members of a district must hold to similar standards and should try to have details of practice conform in so far as possible. The institution acting as clearing house to a large district ought to be centrally located.

D. Cattle clubs. It is hardly conceivable that the cattle clubs would be able to take over the administration of advanced registry work at the present time or in the very near future. Impaired financial condition, internal politics and mutual jealousies, all operate against a change in that direction now. It would seem to be quite feasible for the breed organizations to jointly take over and carry advanced registry testing sometime in the future. The committee on relations may well have this possibility in mind. Meanwhile, the colleges and experiment stations seem to be the most logical as well as competent instruments for the job, so can hardly shift the responsibility.

2. REPORT OF SUB-COMMITTEE ON UNIFORM RULES FOR THE CONDUCT OF OFFICIAL RECORDS OF DAIRY COWS

Rules must be followed. 1. The supervisor is not at liberty to decide as to which stipulations contained herein are essential and which are not, but is required to observe these directions in all details.

Relationship of supervisor. 2. The supervisor shall bear in mind at all times that he is in the employ of the agricultural college or experiment station of the State employing him and that he is not in any way working for the owner of the cow on test or for the breed association. The *only interest* that the supervising institution has in the test is that it shall be honestly and accurately made and the agricultural college or experiment station supervising said test *insists* that this be done.

Identity and condition of cow. 3. The supervisor shall satisfy himself as to the identity of each cow under test. Certificates of registration or authorized official diagrams of color markings must be presented

for the identification. Animals not having distinctive markings indicated on registration papers must be tattooed in the ear with such letters, characters or numbers as the owner may adopt. These identification marks must be recorded with the breed association and the institution as well. All animals which cannot be identified under the above rules must be reported as unidentified. Complete identification is of the utmost importance and *must not be neglected*. The supervisor shall note upon his report form any sickness of a cow or other condition likely to affect the reliability of a test. He shall also report any irregularity or suspicious occurrence.

Unidentified cows. 4. Unidentified cows are allowed to be placed on test, the institution however, assuming no responsibility as to the identity of the animal—this matter must be attended to between the the owner and the breed association. Test reports of unidentified cows must be accompanied by color sketch or description of the cow so tested.

Possessor considered owner. 5. For all purposes of official testing work, the possessor is considered the owner and is required to treat the animal as such, presenting all necessary data, the same as for his own.

Preliminary milkings. 6. Preliminary milkings are required. The supervisor shall be present at the last regular milking preceeding the beginning of the test, and shall see that the cow is milked dry. He shall note the hour that this milking is made, and the last milking of the test shall be made at the same hour.

Seeing the cow milked. 7. The supervisor shall be present at and throughout each and every milking during the test period and shall see that the pail contains nothing but the milk drawn from the cow under test. Before each milking period, the supervisor shall observe that the milk pail is free from grease or other fatty substance and the supervisor shall carry the milk pail at all times during the milking period, allowing the pail out of his hands only after the milker is seated, ready to begin milking, and during the actual milking process. Under no circumstances shall more than one cow undergoing test be milked at the same time and the supervisor must in every case be in position to observe the milker during the entire milking process.

a. Milking machines. Where a milking machine is used, no second man as stripper is allowed. The milking machine is to be run idle for a few moments before attaching to the cow to insure that no milk is coming from another source. Only one cow shall be milked at a time, the same as in hand milking.

b. Right of search. The supervisor has the right to search the milker at any time and to require milkers to roll sleeves up to the elbows. Refusal on the part of the milker will be construed as evidence of intent to make a fraudulent test.

Weighing milk. 8. The supervisor shall assure himself as to the accuracy of the scales used, checking those of the owner with those provided by the institution, these being graduated to pounds and tenths ($\frac{1}{10}$) of a pound. He shall weigh the empty milk pail before each milking, retaining possession of the pail until the milker is seated and ready for milking. After each milking is completed he shall weigh the same immediately and record the exact weight of the milk in his field notes and also see that the correct weight is recorded upon the owner's barn record. The pounds of milk must be recorded in one decimal, as 14.5 pounds. Any inaccuracy in the owner's scale shall be reported to the breed association and to the superintendent of official testing in the State concerned.

Sampling milk. 9. The supervisor shall take a sample of the milk of each milking immediately upon the weighing of the milk, being careful that the milk is thoroughly mixed by pouring from one pail to another at least twice to insure a fair sample of the whole—the owner being required to provide an extra pail for this purpose. Such samples (properly labeled) must be securely retained until tested, under the absolute control of the supervisor, either being in his actual possession or securely locked in the testing case provided. The samples must be tested as soon as convenient after the samples have cooled to ordinary room temperature, which is 60–70°F. When the last samples at night are kept until morning, in warm weather preservatives should be added to each sample.

Applying the Babcock test. 10. The supervisor shall apply the Babcock test to the milk of every milking during the test period, making a separate test in duplicate of each milking. He shall determine the percentage of butter fat in each sample, making a duplicate test of each milking and recording the same. Should the duplicate tests of each sample of milk vary more than two-tenths of one per cent (0.2 per cent), the testing of the sample of milk must be repeated. If the variation is 0.2 per cent or less, then the average of the duplicate tests shall be used in figuring the amount of butter-fat in the sample of milk. Readings of the test bottles shall be made at 130 to 140°F. This temperature shall be obtained by placing the bottles in a hot water bath for five minutes before reading the tests. The supervisor shall record these

readings immediately in his field notes. Samples taken at any one milking shall not be thrown away until satisfactory duplicate tests of the milking are obtained.

Lost milk and lost samples. 11. No substitution of lost milkings or lost samples is allowed. The supervisor shall report that portion of the test for which *exact* data is at hand. Any missing data due to loss of milk weights or test samples are to be left blank on the report.

Report of high production. 12. When the following production requirements in a two day (forty-eight-hour) test period have been exceeded, a special report of the same must be made to the state superintendent of official testing.

	<i>Pounds of fat</i>
Junior 2-year old.....	4.0
Senior 2-year old.....	4.4
Junior 3-year old.....	4.8
Senior 3-year old.....	5.2
Junior 4-year old.....	5.6
Senior 4-year old.....	5.8
Mature.....	6.0

Preliminary reports. 13. A preliminary report card shall be mailed by the supervisor from the farm to the breed association immediately after the test.

Reports of supervisors. 14. The supervisor shall sign his reports and mail them promptly to his appointing officer for checking and endorsement.

Number of milkings allowed to be supervised at one time. 15. No supervisor is allowed to supervise more than (24) twenty-four milkings per day, without special permission from the state superintendent of official testing, unless all cows under test are milked twice daily, when a limit of thirty milkings is permissible.

Test periods exceeding two days. 16. Where a test is conducted longer than the regular two days or where more than one test is made in a month, all details of each test from beginning to end must be reported.

Payment of supervisors. 17. Under no circumstance shall any payment, gift, or gratuity to the supervisor be made by, or permitted from the owner of the cow or any one interested in her and any violation of this rule will invalidate the test.

Supervisors to supervise tests. 18. The supervisor appointed to conduct a test is the direct representative of the Superintendent of Official Testing and needs to be treated in all respects as though he were the appointing officer in person. His duties are to supervise tests only.

Sworn statements. 19. Each supervisor is required to fill out a sworn statement covering all his work which eliminates the necessity of his affirming test reports.

Responsibility for enforcement of rules. 20. Owners, and persons in their employ, are held equally responsible with the supervisor for the enforcement of the foregoing rules.

APPENDIX TO INSTITUTION RULES GOVERNING THE CONDUCT OF
OFFICIAL TESTS

A. Steps in the Babcock test of whole milk

1. Temper sample of milk to 60–70°F.
2. Mix thoroughly by pouring from one bottle to another three or four times—never shaking samples.
3. Pipette 17.6 cc. milk, taking a duplicate test.
4. Add 17.5 cc. sulphuric acid, adding more than 17.5 cc. acid if acid is weak and less if strong. Desired color being a light coffee color.
 - a. Strong acid causes charring of the solids not fat; dark fat columns; specks in the fat column.
 - b. Weak acid leaves undissolved particles at base of fat column; very light colored fat column; grayish or white particles in fat column.
5. Whirl at proper speed for five minutes, usually eighty revolutions of handle per minute.
6. Add hot water (170–180°F.), bringing fat to neck of bottle. Above temperatures apply to hand testers.
7. Whirl two or three minutes.
8. Add hot water (180°F.), bringing the fat column to the 7 per cent mark. Above temperatures apply to hand testers.
9. Whirl one or two minutes.
10. Place water bath of 130–140°F. for five minutes.

Be sure entire fat column is surrounded with the hot water.
11. Read from bottom of fat column to extreme top of fat column, using dividers for this purpose.

B. Recording tests

In recording tests use the following system: The percent of fat to be recorded in one decimal, as 3.2 per cent, except where a difference of 0.1 per cent exists between duplicates, when the percent of fat shall be recorded in two decimals (3.2 per cent–3.3 per cent) 3.25 per cent. The pounds of fat shall be recorded in three decimals, as 0.345 pounds of fat and in case the fourth decimal is five or above, add one to the third decimal, as 0.3456 will give .346 pounds fat.

C. Cautions

1. Label samples and tests of samples so that no possible confusion may ensue.
2. Accept only accurate clear cut fat columns.
3. Watch temperatures—use your thermometer—never guess temperatures.
4. Avoid unnecessary breakage of glassware.
5. Keep glassware scrupulously clean. Clean equipment at the close of every test.

D. Determining fat percentage

The average percentage of fat shall be determined by dividing the total yield of fat for any test period by the total yield of milk for the same test period. It should be recorded in two decimal places as 3.27 per cent. In case the fourth figure in the division is five or above, then add one to the third figure, as 3.267 would give 3.27 per cent.

RECOMMENDATION TO STATE SUPERINTENDENT OF OFFICIAL TESTING

Number of supervisors required. 1. Not less than three (3) supervisors are required on every yearly test.

3. REPORT OF SUB-COMMITTEE ON METHODS OF FINANCING OFFICIAL TESTING

We recommend:

1. The adoption of the flat rate

(1) That each state adopt the flat rate method, that is, make a per day or per test charge that will cover all the charges for test supervision.

(2) That the amount of this charge be decided by each State according to local conditions, such as the local price of labor, number of herds under supervision, the particular system of supervision being used and the amount of travel and hotel charge that may be required in connection with the supervision.

The following are advantages of the flat rate method of charging:

- (1) It equalizes the expense to the breeders. Breeders living at great distance pay no more than breeders living close to the college.
- (2) It greatly simplifies the keeping of advanced registry accounts.
- (3) It avoids misunderstanding with the breeders.
- (4) The breeder knows in advance exactly what it will cost to conduct any given test.

(5) Supervisors knowing the limits to which they are held will be more careful in incurring expenses.

2. Charges should cover the cost of advanced registry supervision

We believe that the department or agency of the college or experiment station, under which the supervision comes is justified in making the charge to the breeder large enough to cover all of the expenses incurred in supervising advanced registry tests as follows:

- (1) Telephone and telegraph.
- (2) Stationary and office supplies.
- (3) Office equipment—supervisor's equipment.
- (4) Salary of superintendent.
- (5) The salaries of all clerical force and assistants necessary in checking and reporting the records.
- (6) Salary or wages of supervisors.
- (7) Travelling expenses of supervisors.
- (8) Notary fees.

3. The breed association should collect from the breeder

(1) A copy of the statement of charges for any given test should be mailed to the cattle club concerned.

(2) That the cattle club shall remit within 30 days to the college.

(3) That a duplicate copy of the statement be kept in the office files.

The advantages of having all bills paid by the cattle clubs are:

(1) It protects the colleges or experiment stations against financial losses.

(2) It removes the necessity of direct financial dealing with the breeder which (a) will eliminate disagreements and (b) enable the college or station to fix adequate charges for financing advanced registry testing.

(3) It simplifies the accounting.

(4) It insures the supervisor of prompt payment in states where they are paid after collections are made from the breeders for whom they tested.

ROY T. HARRIS,
W. W. YAPP,
H. N. COLMAN,
J. B. FITCH,
WM. J. REGAN,
G. C. WHITE,
E. G. WOODWARD,
Committee.

CONFERENCE WITH REPRESENTATIVES OF BREED ASSOCIATIONS

ROY T. HARRIS, *Secretary of the Conference*

This conference of the committee on relation to breed associations of the American Dairy Science Association with representatives of the Breed Associations was held at the Hotel Harrington, Washington, D. C., December 20, 1921.

Those present were: O. H. Baker, A. J. C. C., C. L. Burlingham, A. C. B. A., Wm. H. Caldwell, A. G. C. C.; M. H. Campbell, Illinois; W. W. Fitzpatrick, Clemson College, South Carolina, M. H. Gardner, H. F. Assoc.; R. M. Gow, A. J. C. C.; Roy T. Harris, Wisconsin; W. M. Regan, New Jersey; G. C. White, Connecticut; E. G. Woodward, Washington.

The purpose of the conference was primarily to place the financial phase of test supervision on a more satisfactory basis. This was clearly stated by Chairman Woodward and Mr. Regan. It immediately developed that each representative of a breed association present was there to assist in the solution of problems which have for a long time bothered the superintendents in the various states and which have led to much misunderstanding and friction. The committee opened negotiations in the spirit of seeking to clear up matters for the good of all.

It was voted as the sense of this conference that the financing of the advanced registry and register of merit work is a responsibility of the breed associations and not of the institutions. All were pretty thoroughly agreed on this point.

After considerable discussion of various schemes which proved more or less objectionable or impracticable, Secretary Caldwell announced the action of his executive committee accepting the plan recommended by the American Dairy Science Association and agreeing to place same in effect at once. This was followed by statements from Secretary's Burlingham and Gow promising endeavors to secure similar action at the next meeting of their respective boards in January, 1922. Secretary Gow invited Messrs. Regan and White to present the American Dairy Science

Association plan to the board meeting of the American Jersey Cattle Club. This was agreed to. Superintendent Gardner pointed out some obvious difficulties in the way of the application of this plan to his association, but agreed to present it to his board. This cannot be done, however, before June, 1922. It was agreed by all that the arrangement could only apply to states having a flat rate for tests.

Breed representatives described the unsystematic methods of billing accounts by some institutions and urged that colleges make a very serious effort to arrive at more uniformity and careful attention to such details. It is agreed that when club forms are furnished, we should use them so far as possible.

Some objection developed to including salary of superintendent as legitimate item of expense of supervision, it being urged that if this is done, he becomes an employe of the breed associations, both in law and fact. No action taken as outside jurisdiction of our conference.

Uniform blanks for reporting one and two day tests of semi-official cows are desirable and have long been urged by the American Dairy Science Association. Secretary Gow made a motion expressing the sense of the conference in favor of the desirability of uniform blanks for reporting such tests which was carried without dissent. After some discussion Mr. Baker of the American Jersey Cattle Club and Mr. Regan of New Jersey were appointed as a committee to work out details of forms to be submitted for approval, or blue pencil, to the parties interested and to devise satisfactory means of printing and distributing the forms to the colleges. It is hoped that the new arrangement will be in effect by the beginning of the next testing year.

The rules for control of test supervisors in the field adopted by the American Dairy Science Association at the St. Paul meeting were submitted to the conference for criticism and suggestion. Some changes in wording were made to meet these, but none affect the essential requirements. If these are approved by the executive committee, they will become the official wording. Advanced registry superintendents in the various states

who have not secured a copy of the new rules may do so by addressing the secretary.

It was re-affirmed as the sense of this conference that the breed associations should accept the rules adopted by the American Dairy Science Association as the official test rules and should publish these as such or not publish any. This in so far as the actual supervision of the tests is concerned. The basis for advanced registration and requirements for entry are strickly breed affairs and subject only to suggestion on the part of the institution.

With the view to assist the breeds represented in harmonizing their practice somewhat several points of difference were discussed. Motions were passed favoring a rule that milk records should not begin earlier than the fourth day after calving and official test periods not earlier than the seventh day after calving counting day of calving as the first day. Another stating as the sense of the conference that all advanced registry and registry of merit records be made in one lactation period was also passed.

The conference adjourned confident that real progress had been made and that the institutions and breed organizations have both benefited through the better understanding at which we arrived.

RECOMMENDED METHOD OF HANDLING ADVANCED REGISTRY TESTING ACCOUNTS

The breed association should collect from the breeder.

1. A copy of the statement of charges for any given test should be mailed to the cattle club concerned.

2. That the cattle club shall remit within 30 days to the college.

3. That a duplicate copy of the statement be kept in the office files.

The advantages of having all bills paid by the cattle clubs are:

1. It protects the colleges or experiment stations against financial losses.

2. It removes the necessity of direct financial dealing with the breeder which (a) will eliminate disagreements and (b) enable the colleges or station to fix adequate charges for financing advanced registry testing.

3. It simplifies the accounting.

4. It insures the supervisors of prompt payment in states where they are paid after collections are made from the breeders for whom they tested.

REVISED CONSTITUTION FOR THE AMERICAN DAIRY SCIENCE ASSOCIATION

Article 1. The name of the organization shall be the American Dairy Science Association.

Object

Article 2. The object of the Association shall be to advance the general welfare of the dairy industry, especially the improvement of dairy instruction, by the stimulation of scientific research in all phases of the subject and by improvement in methods of conducting extension work.

Membership

Article 3. Membership shall be of only one kind, namely: Active.

Article 4. The following are eligible for election:

(1) Any person who is formally announced by an agricultural college, or experiment station or by the Dairy Division of the United States or Canadian Departments of Agriculture as an instructor, extension worker, investigator or administrative officer connected with the dairy industry, or anyone filling a position of responsibility connected with the dairy industry and who has had a college or university training in technical science, or anyone filling a responsible position in the industry of a professional character requiring a technical knowledge of dairying of a high order.

Article 5. Nominations for membership shall be submitted to the executive committee in written form and signed by at least three members. Upon receiving the unanimous endorsement of the executive committee and paying the annual membership dues, the candidate shall be duly enrolled as a member of the Association.

Officers

Article 6. The officers of this Association shall be president and vice-president, whose term of office shall be one year; a secretary-treasurer and a *Journal* editor, whose term of office shall be two years. The term of office shall begin January 1.

The president, the vice-president and the secretary-treasurer and the presiding officers of the divisions of the Society shall constitute the executive committee.

Duties of officers and manner of election

Article 7. The duties of the officers shall be those usually pertaining to their respective offices. The executive committee shall pass upon all nominations for membership, all applications for divisions and sections of the Society. It shall have the power to fix the time and place for the annual meeting and shall be authorized to transact such business of the Association as demands attention while the Association is not in session.

Article 8. On or before November 1, the secretary shall mail to each member a blank on which he shall be entitled to express his choice for each office to be filled. This blank shall give the names of those serving in the offices to be filled for five years preceding and a report of the committee on elections which shall suggest the names of two members for each office to be filled. All ballots shall be counted by the secretary and later verified by the president. In case no candidate has a majority by the first ballot, a second ballot shall be sent to the members, giving the names of the three leading candidates for each office and the number of votes received. The candidates receiving the most votes on the second ballot shall be declared elected. In case of tie on the second ballot, the decision shall be made by the executive committee.

Organization of divisions and sections

Article 9. Professional groups based upon geographical considerations to be known as divisions of the Society and to be organized by members of the Association may be authorized by the executive committee when such action shall seem expedient. The officers of the division shall be chairman and such other officers as are provided by the division. The presiding officers of division shall be ex-officio vice-presidents of the Society.

The divisions shall have the right to make by-laws for their own government and which shall not be inconsistent with the constitution and by-laws of the Association.

Membership in divisions of the Society is open only to those regularly elected members of the Society.

Any division may raise or collect funds to be expended for its own purpose.

Article 10. Professional groups based upon specialized interests to be known as sections of the Society and to be formed by not less than 10 active members, may be authorized by the executive committee when considered for the best interests of the Association.

Such sections may elect their own officers and may make any rules for their own guidance not inconsistent with the constitution and by-laws of the Association.

Amendments

Article 11. The constitution may be amended by a two-thirds vote at any regular meeting of the Association; provided the proposed amendments have been submitted to the executive committee in writing not less than 30 days previous to the meeting at which the vote is taken; and provided the proposed amendment is approved by a majority of the executive committee.

Article 12. The executive committee may, at its discretion, submit proposed amendments which have received the approval of the committee to the members of the Association for vote by mail. An affirmative vote of two-thirds of all voting and which shall be not less than a majority of the membership shall be necessary for approval.

By-laws

Article 1. The membership dues shall be \$5.00 a year, payable January 1, each year.

Article 2. The *Journal of Dairy Science* shall not be sent to any member whose dues are not paid by April 11 of the year for which the Membership is held.

Article 3. Any members in arrears for dues for more than one year shall thereby cancel membership but may be restored to membership without any action by the Society by payment of all arrears including dues for the current year.

Article 4. The time and place of the meetings of the Society shall be fixed by the executive committee.

Article 5. A quorum at any meeting for the transaction of business shall consist of not less than 10 per cent of the active members.

Article 6. A committee of five on elections shall be appointed by the president. Said committee shall make a report at the annual meeting in which the names of two members shall be suggested for each office to be filled by mail ballot under the provision of the constitution. In addition thereto, a group of five or more members may suggest other names for consideration but such requests shall be in the hands of the secretary of the Association before November 1.

Article 7. In conducting mail ballots, only those ballots shall be counted which are received by the secretary within four weeks from the date upon which the ballots were mailed by the secretary.

Article 8. Section Meetings. The time and place of meeting for the Sections of the Association shall be fixed by the executive committee. All other arrangements regarding the section meetings shall be made by the officers of the section.

Article 9. These by-laws may be amended at any regularly called meeting by a two-thirds vote of those present.

AMERICAN DAIRY SCIENCE ASSOCIATION

Organization for 1922

President.....C. H. ECKLES, University Farm, St. Paul, Minn.
Vice president.....A. A. BORLAND, State College, Pa.
Secretary-treasurer.....J. B. FITCH, Manhattan, Kans.
Editor.....J. H. FRANDSEN, Lincoln, Nebr.

EASTERN DIVISION

Chairman.....R. C. FISHER, Storrs, Conn.

STANDING COMMITTEES REPORTING TO THE GENERAL MEETING

Membership

J. A. GAMBLE.....Maryland, *Chairman*
H. F. JUDKINS.....Massachusetts
W. H. E. REID.....Missouri
V. D. CHAPPELL.....Oregon

Bacteriological methods for market milk

R. S. BREED.....New York, *Chairman*
L. A. ROGERS.....Dairy Division, U. S. D. A.
J. D. BREW.....New York
E. G. HASTINGS.....Wisconsin
B. W. HAMMER.....Iowa

SECTION 1. DAIRY PRODUCTION

Chairman.....W. M. REGAN, Experiment Station, New Brunswick, N. J.
Secretary.....E. L. ANTHONY, Morgantown, W. Va.

STANDING COMMITTEES

Dairy cattle judging contest

W. W. SWETT.....Missouri, *Chairman*
E. L. ANTHONY.....West Virginia
J. B. FITCH.....Kansas
H. H. WING.....New York
H. H. KILDEE.....Iowa
E. V. ELLINGTON.....Dairy Division, U. S. D. A.
W. M. REGAN.....New Jersey

SECTION 2. DAIRY MANUFACTURES

Chairman.....L. A. ROGERS, Dairy Division, U. S. D. A.
Vice chairman.....C. L. ROADHOUSE, California
Secretary.....H. A. RUEHE, Illinois

STANDING COMMITTEES REPORTING TO SECTION 2

Score card for dairy products and legal standards

J. H. FRANDSEN.....	Nebraska, <i>Chairman</i>
H. B. ELLENBERGER.....	Vermont
S. C. THOMPSON.....	Dairy Division, U. S. D. A.
B. W. HAMMER.....	Iowa
L. A. ROGERS.....	Dairy Division, U. S. D. A.
J. A. GAMBLE.....	Maryland
ERNEST KELLY.....	Dairy Division, U. S. D. A.
W. P. B. LOCKWOOD.....	Massachusetts
W. W. FISKE.....	New York
J. L. SAMMIS.....	Wisconsin
O. F. HUNZIKER.....	Chicago, Ill.
A. C. BAER.....	Oklahoma

Dairy products judging contest

A. W. RUDNICK.....	Iowa, <i>Chairman</i>
J. R. KEITHLEY.....	Minnesota
J. A. GAMBLE.....	Maryland
T. E. WRIGHT.....	South Dakota
R. S. STOLTZ.....	Ohio
V. D. CHAPPELL.....	Oregon
E. S. GUTHRIE.....	New York
S. C. THOMPSON.....	Dairy Division, U. S. D. A.

Official methods for testing milk and cream

O. F. HUNZIKER.....	Chicago, Ill., <i>Chairman</i>
F. W. BOUSKA.....	Chicago, Ill.
H. C. TROY.....	New York
L. A. ROGERS.....	Dairy Division, U. S. D. A.
R. H. SHAW.....	Chicago, Ill.

SECTION 3. EXTENSION

<i>Chairman</i>	E. M. HARMON, Columbia, Mo.
<i>Vice chairman</i>	C. A. HUTTON, Knoxville, Tenn.
<i>Secretary-treasurer</i>	C. R. GEORGE, Lafayette, Ind.

STANDING COMMITTEES REPORTING TO SECTION 3

Cow test association

C. R. GEARHART.....	Kansas, <i>Chairman</i>
E. V. ELLINGTON.....	Dairy Division, U. S. D. A.
M. H. KEENEY.....	New Jersey

Bull associations

A. C. BALTZER.....	Michigan, <i>Chairman</i>
L. A. HIGGINS.....	Mississippi
J. G. WINKLER.....	Dairy Division, U. S. D. A.

Calf clubs

C. A. HUTTON.....	Tennessee, <i>Chairman</i>
E. B. FITTS.....	Oregon
C. B. FINLEY.....	Iowa

Milk campaigns

MISS JESSIE HOOVER.....	Dairy Division, U. S. D. A.
JNO. L. WANN.....	Indiana
R. H. OLNSTEAD.....	Pennsylvania

Dairy products

L. W. MORLEY.....	Pennsylvania, <i>Chairman</i>
W. L. CLEVINGER.....	Tennessee
CARL W. SCHMOLKE.....	South Carolina

SECTION 4. OFFICIAL TESTING

<i>Chairman</i>	H. P. DAVIS, Nebraska
<i>Vice chairman</i>	L. H. FAIRCHILD, Indiana
<i>Secretary</i>	ROY T. HARRIS, Wisconsin

STANDING COMMITTEES

Relation to breed association

G. C. WHITE.....	Connecticut, <i>Chairman</i>
H. M. COLMAN.....	Oregon
J. B. FITCH.....	Kansas
ROY T. HARRIS.....	Wisconsin
W. M. REGAN.....	New Jersey
W. W. YAPP.....	Illinois
A. A. BORLAND.....	Pennsylvania

Methods of investigation

A. C. RAGSDALE.....	Missouri, <i>Chairman</i>
H. H. WING.....	New York
W. E. PETERSON.....	Minnesota

Composite samples

L. H. FAIRCHILD.....	Indiana
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BITTER MILK OF ADVANCED LACTATION¹

A LIPASE FERMENTATION

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Bitter milk is ascribed most commonly to specific bacteria, of which several species have been described by various investigators (1). Less commonly a diseased condition of the udder (2) or the eating of certain weeds, such as rag weed, causes the milk to be bitter. The two latter causes are distinguished from the first in that the milk is bitter when drawn, whereas bitterness due to bacterial fermentations develops after the milk is secreted. Still another type of bitter milk is associated with advanced lactation. This type was described in some detail by Eckles and Shaw (3) in the case of several cows under experimental conditions where the feeding conditions and care and handling of the milk were uniform. As described by these investigators the milk developed a strong rancid odor, suggestive of butyric acid within twenty-four hours after it was drawn from the cow. This odor was accompanied by a very bitter flavor. It was observed that these conditions developed also in the cream separated from the milk. In neither case did the addition of formalin to the fresh products prevent the development of the abnormal flavor and odor. Extreme difficulty of churning the cream also accompanied these conditions.

That the conditions described by the aforementioned writers are not uncommon in the commercial production of milk is indicated by their statement that "numerous inquiries are received each year regarding abnormal flavor in milk from cows near the end of the lactation period." The writer's attention

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was called to the question of the cause of this abnormality because of the numerous inquiries received regarding it. An examination of these inquiries shows also that almost invariably the inquirer has only one or two cows, indicating that the conditions described are not necessarily of great practical importance in dairies or for market milk.

The present paper describes an experimental study of the cause of the so-called bitter milk of advanced lactation. This inquiry has been continued over a period of several years because it has been necessary to study the problem when typical cases were so available that the handling of the samples of milk could be rigidly controlled. Unfortunately for the rapid completion of the study most of the inquiries received were from persons located so that an experimental study of the milk was not possible.

It is believed that sufficient data have now been secured to warrant the conclusion that the immediate cause of the abnormal flavor and odor of milk of certain cows whose period of lactation is more or less advanced is the secretion of an abnormal quantity of lipase in the milk. The proof for this will be furnished below. It will be shown that it is not possible to transfer this condition by inoculation from typical "bitter milk" of this type to normal milk. It will be shown, also, that such milk exhibits an abnormal lipase activity even under bacteriologically sterile conditions. Finally it will be shown that milk which will develop this abnormal flavor and odor can be prevented from so doing by heating to a temperature which is ordinarily regarded as destroying lipase activity.

While the experimental results reported below are believed to show that the immediate cause of the bitter flavor and rancid odor of milk from cows in advanced lactation is an active lipase in the milk, the author has not as yet gone back of this question to ascertain the physiological explanation for the presence of this lipase. Numerous studies which the writer carried out on the normal presence of lipase in cow's milk in connection with this investigation, which were reported in a preceding paper (4),

showed that cow's milk does not normally contain a true lipase. It is generally believed that an active lipogenesis occurs in the mammary gland during lactation. This theory presupposes the presence of a lipase in the udder cells. It is not surprising, therefore, that this enzyme should, at times, occur in the milk. In fact, it is surprising that an active lipase does not occur in milk normally. Inasmuch as lipase is apparently absent under normal conditions, the physiological problem to be solved in the case of the bitter milk of advanced lactation is the factor underlying its appearance at this particular time. It is clear that the conditions causing its appearance do not occur for all cows at the close of lactation. Advanced lactation, therefore, is not in itself the primary cause. Obviously the whole matter is much more deep seated, and statements which may be advanced to explain the phenomenon can only be speculative. The problem is worthy of further investigations, which the author hopes later to undertake.

One of the striking phenomena which frequently accompanies the abnormal fermentation described is the extreme difficulty of churning the cream from the milk. This condition was pointed out by Eckles and Shaw (3) and inquiries as to a remedy for the difficulty often accompany requests for information regarding bitter milk. The principal characteristic of the cream in these cases is excessive foaming. This, of course, is the primary cause of the difficult churning. It was observed in the course of these studies that the milk itself also foams very readily when shaken and that this property increases as the milk becomes more rancid. This observation indicates that the difficulty of churning is directly connected with the fermentation which causes the rancid odor and bitter flavor and is no doubt caused by the formation of soaps which accompanied the liberation of free fatty acids during the lipolytic fermentation. It is recognized, also, that this cause of the difficult churning is augmented by the conditions which usually render churning more difficult at the close of lactation, namely, higher protein content and smaller fat globules.

EXPERIMENTAL

Bacteria as possible cause of abnormal fermentation

Three cases of bitter, rancid milk were examined particularly with the view of determining whether the abnormal fermentation could be transmitted from the bitter product to fresh, sweet milk or cream. In each case only a single cow was involved and the milk and cream showed the characteristics mentioned to a marked degree within ten or twelve hours after milking, even when kept cold. Great difficulty of churning accompanied the conditions in one case. Observations regarding this difficulty were not made in the other two cases inasmuch as the milk was used only for consumption as whole milk and cream.

The procedure for examining the transmission of the fermentation was essentially the same in each case and consisted of inoculating separate samples of freshly pasteurized sweet cream with increasing amounts of the bitter, rancid product, beginning with 1 drop and ending with 5 cc. to 7 cc. In some cases 15 cc. of the sweet cream were used and in other 17.6 cc. In all cases the inoculated material was kept in 50 cc. Erlenmeyer flasks, stoppered with a cotton plug. Observations as to odor and flavor were made at intervals of twenty-four and forty-eight hours after keeping the samples at room temperature or 38°C. In the tests with one sample of rancid milk a set of samples was kept at 5-8°C., also, and one sample was incubated at 38°C. in which 7 cc. of the bitter rancid milk were heated to 72°C. before adding it to 17.6 cc. of sweet cream.

It does not seem necessary to report the protocols of the several tests inasmuch as the results were uniform throughout. Inoculations with 1 or 2 drops uniformly failed to produce the rancid odor and bitter flavor under each of the temperatures employed. When 5 cc. or more of the rancid milk were added to 15 cc. to 17 cc. of sweet cream more or less of the characteristics of the original product developed on standing. The most striking results were secured in all cases when the largest amount (7 cc.) of the rancid material was added and when the temperature of the mixture was maintained at 38°C.

The results of these tests indicated clearly that the origin of the fermentation is not bacterial but enzymatic since the characteristics of the original product fail to develop on dilution of the causative factor and also fail to develop on inoculation with amounts which would cause the fermentation if bacteria were responsible. In the single case where the original product was added in large amount (7 cc.) after heating to 72°C., the fermentation also failed to develop, even at 38°C.

Attention should be called also to the fact that the rancid odor and bitter flavor of each of the original products used for inoculation of the sweet cream were strongly suggestive of the development of butyric acid. In fact, when fresh milk was treated with a small amount of an aqueous solution of butyric acid the milk become so nearly identical in flavor and odor with the naturally rancid milk that several observers found it impossible to distinguish between the natural and artificial products. It was this observation, especially, together with the results of the inoculation tests, which led to an examination of the lipolytic activity of the subsequent samples of bitter milk secured for investigation.

Lipolytic activity of bitter milk

The study of the lipolytic activity of the bitter milk of advanced lactation was conducted with three samples secured at separate times. One of these was the third case reported above for a possible bacteriological origin of the fermentation. In each case a single cow, only, was involved. The experiments on this phase of the problem will be reported separately.

Experiment 1. The milk was received from the owner of the cow in two portions. One portion was raw milk to which 1.5 per cent chloroform had been added to the fresh milk by the owner. The other portion had not been so treated but had been heated while fresh to 75°C. Both portions were about seventy hours old when the experiment was begun. At this time the portion treated with chloroform had the characteristic rancid odor and bitter taste in spite of the chloroform present. The heated portion showed neither of these properties.

Four 50 cc. portions of the raw milk were placed in 200 cc. Erlenmeyer flasks and 25 cc. of freshly boiled and cooled distilled water added to each flask, together with 1 cc. of neutral, saturated potassium oxalate solution. Two of these flasks were then heated to 78°C. and cooled at once. Two similar flasks were prepared containing the milk which had been heated by the owner of the cow. 1 cc. of chloroform was now added to flasks 1 and 2, containing the raw milk, and 1.5 cc. to the remaining 4 flasks. After adding 5 drops of 1 per cent alcoholic phenolphthalein solution to each flask all were titrated to a faint pink with 0.1 N aqueous NaOH solution. The titrations were repeated after 2 successive twenty-four hour intervals, the flasks being kept at 37°C. The results are shown in table 1, after calculating to 100 cc. of milk.

TABLE 1
Lipolytic activity of bitter, rancid milk, experiment 1

FLASK	TREATMENT OF MILK	INITIAL ACIDITY*	ACIDITY* AFTER 24 HOURS	ACIDITY* AFTER 48 HOURS
		cc.	cc.	cc.
1	{CHCl ₃ added when fresh.....	33.2	5.60	2.50
2	{CHCl ₃ added when fresh.....	32.6	5.84	2.70
3	{CHCl ₃ added when fresh.....	31.4	2.76	1.20
4	{Heated to 72°C. after 70 hours.....	32.0	2.30	1.24
5	{Heated to 72°C. when fresh.....	14.4	3.00	1.76
6	{CHCl ₃ added after 70 hours.....	14.4	3.10	1.60

* Co. 0.1 N acid per 100 cc. milk.

The striking features of the data shown in table 1 are first, the high initial acidity of the milk in flasks 1 to 4 to which a bacterial antiseptic had been added which does not inhibit lipase activity² in comparison with the initial acidity of the milk in flasks 5 and 6 which had been heated when fresh to the thermal death point of lipase; second, the much greater development of acidity in flasks 1 and 2 which were not treated further before incubation in comparison with that which developed in the remaining (flasks 3 and 4) or when fresh (flasks 5 and 6). The results clearly suggest a marked lipolytic activity in this milk.

² Studies on lipase as affected by CHCl₃, subsequently undertaken and recently reported elsewhere (Jour. Am. Chem. Soc.) by the author show that CHCl₃ does retard the activity of lipase. These samples therefore would have shown a still higher initial acidity and greater development of acid had a better antiseptic, such as HCHO, been used.

Experiment 2. In this experiment the milk used was about forty-eight hours old when received. It was strongly rancid and bitter but had not been treated in any way previous to the beginning of the tests. The technic employed was the same as in experiment 1, except that a set of flasks was prepared for study in conjunction with the bitter milk, using fresh normal milk from the University herd. The following flasks were prepared:

Bitter milk

Flask 1. 50 cc. milk + 0.5 cc. CHCl_3 .

Flask 2. 50 cc. milk + 0.06 cc. formalin.

Flask 3. Same as 1 except heated to 72°C . for thirty seconds.

Flask 4. Same as 2 except heated to 72°C . for thirty seconds.

Normal milk

Flask 5. 50 cc. milk + 0.5 cc. CHCl_3 .

Flask 6. 50 cc. milk + 0.06 cc. formalin.

Flask 7. Same as 5 except heated to 72°C . for thirty seconds.

Flask 8. Same as 6 except heated to 72°C . for thirty seconds.

The results of this experiment are shown in table 2, and are summarized in table 3, the data being calculated in terms of 100 cc. of milk.

The data, like those obtained in experiment 1, show that the bitter, rancid milk of advanced lactation exhibits a pronounced lipolytic activity in comparison with normal milk and in comparison with the bitter milk which has been heated to the thermal death point of lipase. The activity of the lipase is seen to be decidedly greater in the presence of formaldehyde than in the presence of chloroform. This result is in conformity with those reported elsewhere by the author.²

It may be stated incidentally that the milk used in this experiment was employed also for one of the studies in which it was attempted, unsuccessfully, to transmit the bitter fermentation to normal milk by gross bacterial inoculation.

Experiment 3. In this experiment the milk used was only three or four hours old. It had a slightly bitter taste but the rancid odor had not yet developed.

The method of determining the activity of lipase in this milk was essentially the same as that used in the two preceding experiments but the technic employed was modified to secure the maximum results, in conformity with studies on lipase published elsewhere by the author (5).

The chief features of this technic are to use sufficiently large samples to permit the taking of aliquots for acidity determination and the use of alcoholic KOH and sufficient fat solvent to insure the determination of all the free fatty acids liberated in the hydrolysis.

TABLE 2

Lipolytic activity of bitter, rancid milk in comparison with normal milk, experiment 2

FLASK	INITIAL ACIDITY*	ACIDITY* AFTER 24 HOURS	ACIDITY* AFTER 48 HOURS
	cc.	cc.	cc.
1	31.84	10.00	4.20
2	38.40	14.44	9.60
3	29.35	3.58	2.30
4	27.30	3.00	2.00
5	23.30	4.48	2.60
6	30.00	4.56	6.10
7	23.10	4.80	2.80
8	29.12	3.60	2.80

* Cc. 0.1 N acid per 100 cc. milk.

TABLE 3

Summary of lipolytic activity of bitter milk, experiment 2

FLASK	ANTISEPTIC	0.1 N ACID DEVELOPED IN 100 CC. MILK	
		In 24 hours	In 48 hours
Bitter milk			
		cc.	cc.
1	1 per cent CHCl_3	10.00	14.20
3	1 per cent CHCl_3 + heat.....	3.58	5.88
2	HCHO 1:2000.....	14.44	24.04
4	HCHO 1:2000 + heat.....	3.00	5.00
Normal milk			
		cc.	cc.
5	1 per cent CHCl_3	4.48	7.08
7	1 per cent CHCl_3 + heat.....	4.80	7.60
6	HCHO 1:2000.....	4.56	10.66
8	HCHO 1:2000 + heat.....	3.60	6.40

The following flasks were prepared:

Flasks 1 and 2. 150 cc. milk + HCHO, 1:1500.

Flasks 3. 225 cc. milk, heated in water bath at 63°C. for thirty minutes, cooled to 20°C., 100 cc. withdrawn, and HCHO 1:1500 added to remainder.

Flask 4. 225 cc. milk, heated to 72°C. for two minutes, cooled to 20°C., 100 cc. withdrawn, and HCHO 1:1500 added to remainder.

Flask 5. 225 cc. milk, heated just to boiling, cooled to 20°C., 100 cc. withdrawn, and HCHO 1:1500 added to remainder.

Flask 6. Portion withdrawn from flask 3.

Flask 7. Portion withdrawn from flask 4.

Flask 8. Portion withdrawn from flask 5.

Flasks 6, 7, and 8, which contained no antiseptic, were stoppered tightly with cotton and placed in the refrigerator at 10°C. to determine if rancidity would develop. Flasks 1 to 5 were used for the lipase determinations as follows: 25 cc. of the milk were withdrawn with a pipette, 1 cc. of saturated potassium oxalate added and the mixture brought to a faint pink with 0.1 N alcoholic KOH, using 5 drops of 1 per cent alcoholic

TABLE 4
Lipolytic activity of bitter, rancid milk, experiment 3

FLASK	TREATMENT	0.1 N ACID DEVELOPED IN 100 CC. MILK IN 24 HOURS
		cc.
1	HCHO 1:1500 only.....	23.77
2	HCHO 1:1500 only.....	24.76
3	Heated to 63°C. for 30 minutes + HCHO 1:1500.....	12.12
4	Heated to 72°C. for 2 minutes + HCHO 1:1500.....	1.70
5	Heated to boiling + HCHO 1:1500.....	0.23

phenolphthalein as indicator. One hundred cc. of neutral ethyl alcohol and then 100 cc. of neutral ether were now added and after thorough shaking the mixture was again brought to a final end point with the alkali. Similar titrations were made on 25 cc. aliquots withdrawn after twenty-four hours' incubation at 37°C.

The results of these determinations are shown in table 4, the data secured being calculated to 100 cc. milk. Inspection of the table indicates clearly a marked acid fermentation in the case of the samples treated with formaldehyde, which was retarded appreciably when the milk was heated in accordance with the usual pasteurization temperatures and which was practically inhibited when the milk was heated to the thermal death point of lipase or above.

The examination of the portions of the heated samples placed in the refrigerator without addition of formaldehyde showed that samples 7

and 8 heated to 72°C. or above failed to develop the bitter flavor or rancid odor after forty-eight hours in the refrigerator, while sample 6, heated only to 63°C. developed these characteristics slightly after twenty-four hours and distinctly after forty-eight hours in the refrigerator. Even at this time, however, the condition of this sample was good in comparison with the remainder of the unheated samples 1 and 2 which had been incubated for twenty-four hours at 37°C. without any previous heat treatment. The odor of butyric acid was very strong in these samples.

DISCUSSION OF RESULTS

The so-called bitter milk of advanced lactation is probably not a matter of great practical importance. In view, however, of the evidence presented in this paper that an active lipase secreted in the milk is the cause of the difficulty, the possibility is suggested that the very rancid butter which one at times encounters, especially from the owners of only one or two cows, may be caused by a lipase of this origin. The author hopes to ascertain, when opportunity presents itself, how high a contamination of normal cream with cream from a cow giving milk of the character under discussion is required in order to affect seriously the market quality of the butter.

One of the features of the inquiries which are received in connection with bitter milk of physiological origin is how the milk in question can be made fit for use. In view of the fact that there is no reason to believe that such milk is unhealthful, the solution of this phase of the problem seems to lie in the success secured in the experiments reported above in preventing the development of the bitter, rancid characteristics by heating the milk to about 75°C. for a few minutes. This procedure should not impair the milk in any way for any of its normal uses, and, from the results obtained, should effectively prevent the development of the objectionable characteristics for the period during which the milk would ordinarily be kept.

CONCLUSIONS

Bitter milk of advanced lactation is caused by the secretion of an active lipase in the milk which hydrolyzes the milk fat quite rapidly, even at fairly low temperatures, with the libera-

tion of fatty acids among which are the lower, volatile fatty acids, especially butyric, which imparts, in large measure, the bitter flavor and rancid odor to the milk. The difficulty is not of bacterial origin. It can be effectively retarded, if not prevented entirely, by heating the fresh milk to 75°C. for a few minutes.

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THE EFFECT OF TEMPERATURE ON THE PERCENTAGE OF FAT IN MILK: A FIRST REPORT

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It is, in general, known that there is a relation between the time of the year, or season, and the percentage of fat in milk. Hills (1) studying the percentage of fat in milk during different periods of the year concludes, "that the tendency of a cow is to give from day to day richer milk when the temperature falls, and poorer as it rises," implying by this statement that temperature is the causative factor. Eckles (2) who found striking variations of the percentage of fat with season which is independent of the period of lactation or of the character of the diet suggests exercise and temperature as possible causes of this variation. Armsby (3) quotes Spier that between 41° and 53°F. temperature fluctuations have no appreciable effect on the percentage of fat. White and Judkins (4) have "no doubt that cold weather is a time for high tests, and warm weather for low ones." Clothier (5) apparently ignorant of Eckles' very important study in which the factor of variability of diet was eliminated, finds it "impossible to believe that the seasonal variation in butter fat content of milk observed in Arizona are not due directly to change of feed."

On a priori consideration, as judged by analogy from the profound influence of temperature on the life processes which have been studied (6) it would be predicted that temperature probably has a directing influence on the percentage of fat (as well as on the milk yield). The close association between season and temperature, and the known relation between season and the percentage of fat, as well as the finding of Hills (1) and the opinion of Eckles (2) that temperature may be responsible for the variations in fat, further encouraged the prediction, and made it seem decidedly worth while testing it out by direct observation between the fluctuation of the environmental temperature and fluctuation of the percentage of fat in the milk.

Accordingly, a group of ten cows at our disposal were set aside for a preliminary observation during the months of March and April of this year. All the conditions under control such as feed and exercise were kept approximately uniform throughout the period, the object being to record data showing the relation of temperature to the percentage of fat in milk uninfluenced, as far as possible under the usual conditions, by other factors. The

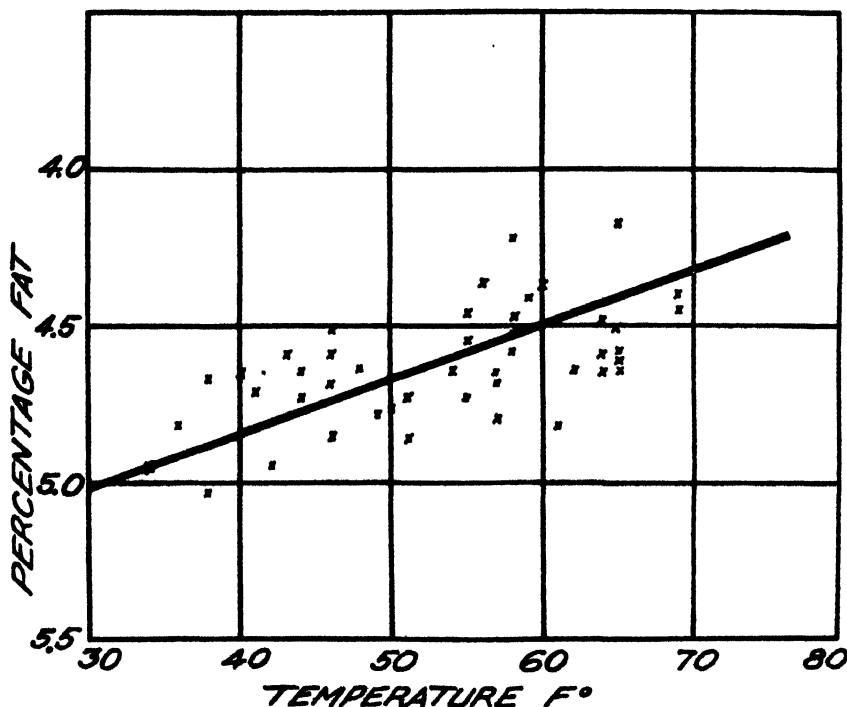


FIG. 1. THE VARIATION IN THE PERCENTAGE OF FAT IN MILK WITH TEMPERATURE.

Ordinates represent the percentage of fat; abscissae represent the environmental temperature in degrees Fahrenheit.

results shown in figure 1 and table 1 in which the average percentage of fat of the group for twenty-four hour periods is recorded against the average temperature of the corresponding twenty-four periods show fairly conclusively, as far as the preliminary experiment is concerned, that there is a relation between temperature and the percentage of fat showing roughly an increase of about 0.2 per cent in the fat for a decrease of 10°F. in

TABLE 1

DATE	MEAN TEMPERA- TURE FOR 24 HOURS	TRUE AVERAGE FOR 24 HOURS
		<i>per cent</i>
March 13.....	57	4.51
March 14.....	60	4.57
March 15.....	60	4.37
March 16.....	47	4.64
March 17.....	58	4.22
March 18.....	64	4.17
March 19.....	70	4.40
March 20.....	69	4.45
March 21.....	41	4.94
March 22.....	43	4.58
March 23.....	48	4.78
March 24.....	55	4.72
March 25.....	58	4.41
March 26.....	64	4.48
March 27.....	45	4.65
March 28.....	27	4.84
March 29.....	39	4.81
March 30.....	44	4.74
March 31.....	41	4.65
April 1.....	46	4.51
April 2.....	49	4.58
April 3.....	64	4.65
April 4.....	65	4.58
April 5.....	65	4.67
April 6.....	64	4.60
April 7.....	61	4.83
April 8.....	55	4.55
April 9.....	39	5.14
April 10.....	41	4.71
April 11.....	46	4.69
April 12.....	59	4.65
April 13.....	56	4.36
April 14.....	56	4.82
April 15.....	62	4.63
April 16.....	37	4.66
April 17.....	37	4.95
April 18.....	45	4.60
April 19.....	57	4.68
April 20.....	58	4.65
April 21.....	60	4.60
April 22.....	55	4.46

the temperature between the observed temperature limits. It is noted in this place that since the experiment was considered as preliminary it was thought permissible to record the percentage of fat against the outdoor temperature, though the animals were kept indoors during stormy days and cooler nights. The barn in which the animals were kept during these stormy and cold periods was well ventilated and not heated artificially.

The practical interest of a knowledge of the effect of temperature on the percentage of fat, for example in connection with official testing, is understood by the dairyman, and need not be discussed. The purely scientific interest will similarly be clear to one who looks over the monographs by Kanitz (6) and Rubner (6). Theories naturally suggest themselves on the possible mechanism connecting the environmental temperature and the percentage of fat, but it is perhaps best to postpone discussion of possible mechanisms until more experimental evidence is available.

SUMMARY

It is shown that all other conditions being approximately the same, the lower the environmental temperature within the observed limits, the higher the percentage of fat in cows' milk.

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BACTERIAL CONTENT OF MILK POWDER

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Aside from the attention which has been given to the question of whether or not living pathogenic bacteria, notably the tubercle organism, may be present in milk powder, there has been but comparatively little information recorded. The particular interest in the work reported by Delepine (1) is centered upon the results involving the virulence of the tubercle organism after being subjected to the temperatures of drying by the hot cylinder process. These results briefly indicate that the tubercle organism of bovine origin is rendered harmless after exposure to the temperatures used for desiccating milk by the Just hot cylinder process. In the same report references to other investigations are cited, the results from which seem to agree that infection by the tubercle organism from desiccated milk made by this process is highly improbable.

Other experiments carried out by Delepine (1) and others involve such matters as the survival of certain non-pathogenic bacteria; the number of bacteria in the powder immediately after drying; its subsequent contamination; and the longevity during storage of those normally present in the finished produce. The results of these experiments show that under normal conditions the number of bacteria per gram of powder as it leaves the drying cylinders is very low, usually only a few hundred, but that recontamination is easily brought about by subsequent handling. The extent of this recontamination under the conditions studied was sufficient to increase the bacterial content from a few hundred to several thousand per gram in many instances.

Downs (2) in an investigation as yet unpublished, found a wide variation in the number of bacteria per gram in milk powder

made by different methods as well as in individual samples from the same method. The average count of ten samples dried by a single roll process (modified Kunick process) in which the milk is previously condensed was 49,500 per gram, with a maximum count of 626,000 and a minimum count of 16,900; all counts except three were below 50,000. The average of ten samples dried by the spray process in which the milk is previously pasteurized and condensed was 178,000 per gram, with a maximum count of 595,000 and a minimum count of 15,600; four counts were below 50,000. The average of nine samples of spray process powder in which the milk is not previously condensed was 2,269,000 per gram, with a maximum count of 3,500,000 and a minimum count of 1,500,000. The counts of these powders were more uniform and much higher than the counts from any other samples thus far reported.

Studies on the types of microorganisms found in commercial milk powder reveal the presence of a large percentage of spore-producing bacteria of the *B. mesentericus* and of the *B. subtilis* types in practically all samples. White and orange coccus forms are also frequently found, as well as yeasts and a variety of molds. From the investigation reported by Delepine (1) it is concluded that the majority of the spore-bearing species survive the heat of the drying process, particularly in the case of the Just double roller process, and that the non-spore-bearing forms are present in the powder as the result of contamination after the powder leaves the drying cylinders.

EXPERIMENTAL

Although it is quite probable that the bacterial content of milk powder is not an important consideration in its sanitary and keeping qualities, it has, nevertheless, been desirable to obtain certain information upon which interpretations as to normal and abnormal conditions with respect to bacterial content of milk dried by the Just hot cylinder process, could be based. While part of the investigation recorded herein is similar in certain features to the work of another investigator to which

reference has already been made, its particular object was to secure data on the longevity of bacteria in milk powder containing various percentages of moisture.

The number and sources of bacteria in dried milk manufactured by the Just process

In order to ascertain the normal bacterial content of milk powder made by the Just process as soon as it is ready for the market, several samples, taken in such a manner as to be representative of the entire day's production, were obtained from different factories and analyzed as soon as possible—usually within one day after manufacture. The bacterial content of these fresh commercial powders is shown in table 1.

TABLE 1
Bacterial content of milk powder made by the Just double roller process

FACTORY	NUMBER OF SAMPLES	MAXIMUM COUNT	MINIMUM COUNT	AVERAGE COUNT
		<i>bacteria per gram</i>	<i>bacteria per gram</i>	<i>bacteria per gram</i>
1	10	10,000	700	4,600
2	26	170,000	2,000	27,300
3	34	96,000	700	18,800
4	13	37,000	800	5,000
5	28	6,360	350	1,500
6	22	8,600	2,100	3,400
7	25	1,400	400	790

The bacterial counts shown in table 1 from milk powders made by the same process at different factories are indicative of wide variations in bacterial quality of milk dried or in the extent of contamination subsequent to the actual drying process. In order to ascertain the extent to which these conclusions may be applicable, liquid milk with a normal flora but with wide variations in bacterial content was dried in the normal manner and the bacteria per gram of powder determined in samples of the dried solids taken immediately after leaving the drying cylinders thus avoiding all possibility of contamination by sifting, handling and packing. The results of these determinations given in table 2 show that the number of bacteria which survive the drying

process is very low, and that with liquid milk containing a normal flora, regardless of the number present, the number which survive is quite constant and does not bear any direct relationship to the number present in the liquid milk before drying. It would seem therefore, that the figures for the normal products (table 1) are largely due to recontamination after drying.

TABLE 2
Bacterial content of liquid milk and powder made from it

SAMPLE NUMBER	LIQUID MILK	POWDER FROM CYLINDER
	<i>bacteria per cubic centimeter</i>	<i>bacteria per gram</i>
1	2, 750, 000	270
2	12, 000, 000	550
3	147, 000, 000	600
4	280, 000, 000	880
5	345, 000, 000	520
6	1, 520, 000	520
7	2, 300, 000	500
8	3, 100, 000	200
9	1, 700, 000	1, 350
10	2, 100, 000	220

TABLE 3
Bacterial contamination of milk powder after drying

SAMPLE NUMBER	MILK SOLIDS FROM CYLINDER	AFTER SIFTING	AFTER PACKING
	<i>bacteria per gram</i>	<i>bacteria per gram</i>	<i>bacteria per gram</i>
1	400	1400	2300
2	960	1400	2300
3	1100	3000	4300
4	600	2200	5200
5	290	1500	4100
6	540	1500	4000
7	50	300	500
Average.....	563	1614	3271

An illustration of the extent of contamination from the drying cylinders to the final package is shown by the results in table 3 in which is given the bacteria per gram of powder at the time the milk solids leave the drying cylinders, after sifting or bolting and after packing. These comparisons were carried out under practical routine factory conditions and illustrate the extent of re-

contamination as a factor in contributing to the total bacteria count of powders made by this process.

The extent to which recontamination can be controlled by proper precautions is well illustrated by the figures in table 4 which show the monthly average bacterial content of the product from one factory before and after thorough provision was made for reducing recontamination to the minimum.

It seems apparent that if the bacterial content of milk powder dried by the Just process is very high, it may be considered as an index of the degree of recontamination after the drying process itself is completed. This statement, however, does not

TABLE 4

Average count of daily samples of milk powder showing high efficiency in controlling bacterial content

MONTH	AVERAGE NUMBER BACTERIA PER GRAM
January.....	1785
February.....	1143
March.....	1195
April.....	800
May.....	650
June.....	405
July.....	590
August.....	584
September.....	595
October.....	600

apply with equal force to the powders made by the spray processes in which undoubtedly a greater number of organisms survive the drying temperatures.

Death-rate of bacteria in milk powder.

Even though the bacterial content of milk powder may be very high, any detrimental effect upon keeping quality is without doubt, eliminated because of the lack of sufficient moisture to allow multiplication. In fact there is every reason to believe that the bacteria which are present in the freshly made product die off rapidly during storage. Since there is no record of suffi

cient data from which a death-rate curve might be constructed, it has been considered desirable to obtain sufficient information for the completion of such a curve.

In order that the death-rate of the organisms in normal powder and in powder artificially inoculated, each with variable moisture content, might be determined, periodic examinations were made on a number of samples extending over a period of one year. Observations were also made to detect possible differences in keeping quality of those samples which were artificially inoculated to the extent of increasing the bacterial content far in excess of that normally found in commercial powders made by the Just process. Samples with variable moisture content were selected for the purpose of determining possible differences in death-rate in those powders containing moisture within the range of variation in this constituent which might possibly exist in the commercial product. Table 5 shows the bacterial content of several of these samples of powder determined immediately after manufacture and at two-month intervals for a period of one year. From these results it is apparent that a point of approximate constancy is reached after two to four months in samples having an initial count of 10,000 or less per gram, whereas a point of approximate constancy is not reached until after six months in the majority of samples originally containing over 10,000 per gram. The death-rate curves constructed from the average of all samples, from the average of samples with the least moisture content and from the average of samples with the highest moisture content are shown in charts A, B, and C, respectively.

The results from powder artificially inoculated are shown in table 6. These results are practically parallel to those from powders with a normal bacterial content in respect to death-rate in high and low moisture samples, although probably due to high initial count, the point of approximate constancy in numbers is not reached as quickly as in the normal samples. The death-rate curve constructed from the average of all samples is shown in chart D.

Examination of the samples recorded in tables 5 and 6 along with check samples containing the same amount of moisture

TABLE 5

Bacterial content of milk powder immediately after manufacture and after storage

SERIES	SAMPLE	MOIST- URE	FRESH	2 MONTHS	4 MONTHS	6 MONTHS	8 MONTHS	10 MONTHS	12 MONTHS
		per cent							
I	1	3.16	3,200	500	400	400	300	300	200
	2	3.94	2,500	600	600	400	400	150	150
	3	5.13	3,300	450	350	350	400	300	200
	4	6.79	2,000	600	300	400	350	400	100
	5	9.34	2,500	1,100	300	400	200	200	100
II	1	3.60	22,000	9,200	1,000	850	150	200	250
	2	4.45	12,500	2,400	1,600	750	500	300	300
	3	5.69	20,000	3,200	2,000	650	350	250	250
	4	6.33	14,000	2,800	3,000	950	300	300	250
	5	9.01	15,000	1,600	700	800	200	300	350
III	1	2.00	750	900	300	350	150	150	200
	2	3.11	1,500	500	300	400	350	250	100
	3	5.05	60,000	55,000	25,000	2,000	350	100	100
	4	5.53	1,400	700	700	500	150	200	200
	5	6.29	100,000	42,000	4,000	900	250	200	50
IV	1	1.74	13,400	8,000	3,500	700	300	250	350
	2	2.72	62,000	15,000	3,500	600	300	300	300
	3	4.02	70,000	41,500	4,000	800	250	200	250
	4	4.33	62,000	15,500	3,000	1,600	350	300	400
	5	5.11	75,000	54,000	5,000	900	300	200	200
V	1	2.27	1,850	1,500	650	700	700	300	250
	2	3.74	17,500	14,000	1,000	400	500	500	400
	3	6.46	80,400	5,000	3,000	600	500	700	400
	4	7.17	75,000	1,000	350	400	500	500	200
	5	7.73	26,500	1,250	1,000	1,000	550	550	450
VI	1	1.91	1,300	500	650	700	700	500	500
	2	3.53	1,350	900	650	800	800	600	600
	3	4.31	2,200	900	900	900	900	550	400
	4	5.51	1,800	800	750	600	600	600	650
	5	6.22	4,200	800	700	700	700	800	550
VII	1	2.50	1,700	250	350	350	250	250	50
	2	4.15	1,100	100	300	300	200	400	50
	3	4.72	3,100	200	250	250	100	200	150
	4	5.20	1,600	200	300	400	300	150	100

TABLE 5—Continued

SERIES	SAMPLE	MOIS- TURE	FRESH	2 MONTHS	4 MONTHS	6 MONTHS	8 MONTHS	10 MONTHS	12 MONTHS
		<i>per cent</i>							
VIII	1	3.64	8,000	1,700	1,700	800	550	200	300
	2	4.92	6,000	650	500	300	450	550	150
	3	5.84	10,200	3,400	1,200	800	700	350	150
	4	6.41	32,000	12,500	2,000	900	850	600	300
	5	7.73	34,000	1,000	650	500	350	350	250
IX	1	3.01	20,000	15,000	15,000	750	350	350	
	2	4.85		5,000	5,500	450	450	450	
	3	5.14		6,000	6,000	400	400	400	
	4	6.76	50,000	8,000	4,000	500	450	500	
Average all samples		4.91	22,508	7,818	2,487	655	412	354	261
Average lowest moisture samples in each series.		2.65	8,022	4,172	2,616	622	383	277	262
Average highest moisture samples in each series.		7.04	34,533	12,216	1,850	677	366	361	256

TABLE 6

Bacterial content of milk powder after artificial inoculation and after storage

SAMPLE	MOIS- TURE	BACTERIA PER GRAM						
		Fresh	2 months	4 months	6 months	8 months	10 months	12 months
	<i>per cent</i>							
1	2.36	730,000	253,000	90,000	95,000	1,050	1,200	850
2	3.84	400,000	8,000	5,000	3,500	1,500	1,200	600
3	4.15	821,000	237,000	40,000	2,000	700	700	700
4	5.37	530,000	43,000	40,000	5,500	3,500	3,800	1,250
5	6.58	1,200,000	440,000	32,000	12,500	12,500	12,000	5,000
6	7.52	1,270,000	244,000	22,000	12,500	12,500	11,000	4,500
Average.....		825,000	201,000	38,000	21,800	5,300	5,000	2,100

but with only a few bacteria per gram did not reveal any differences in quality which could be interpreted as due to the presence of excessive numbers of bacteria.

CHART "A"
Death-rate of Bacteria in Milk Powder.
Average Moisture 4.91%

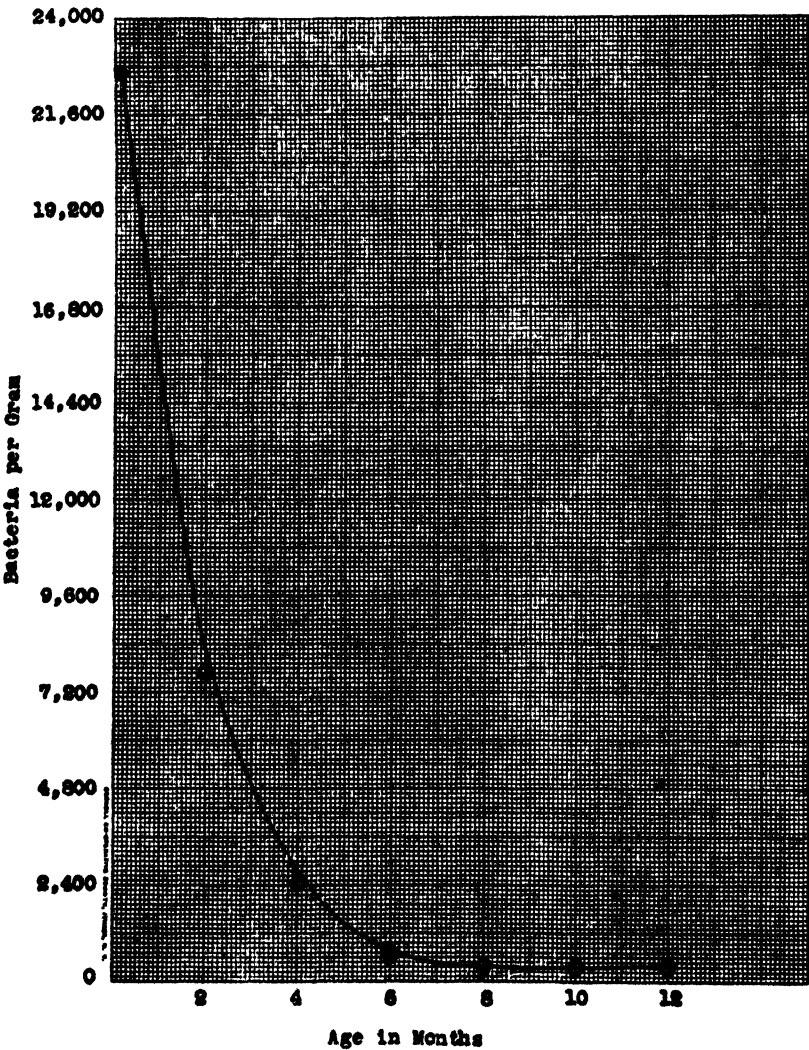


CHART "B"
Death-rate of Bacteria in Milk Powder
Average Moisture 2.65%

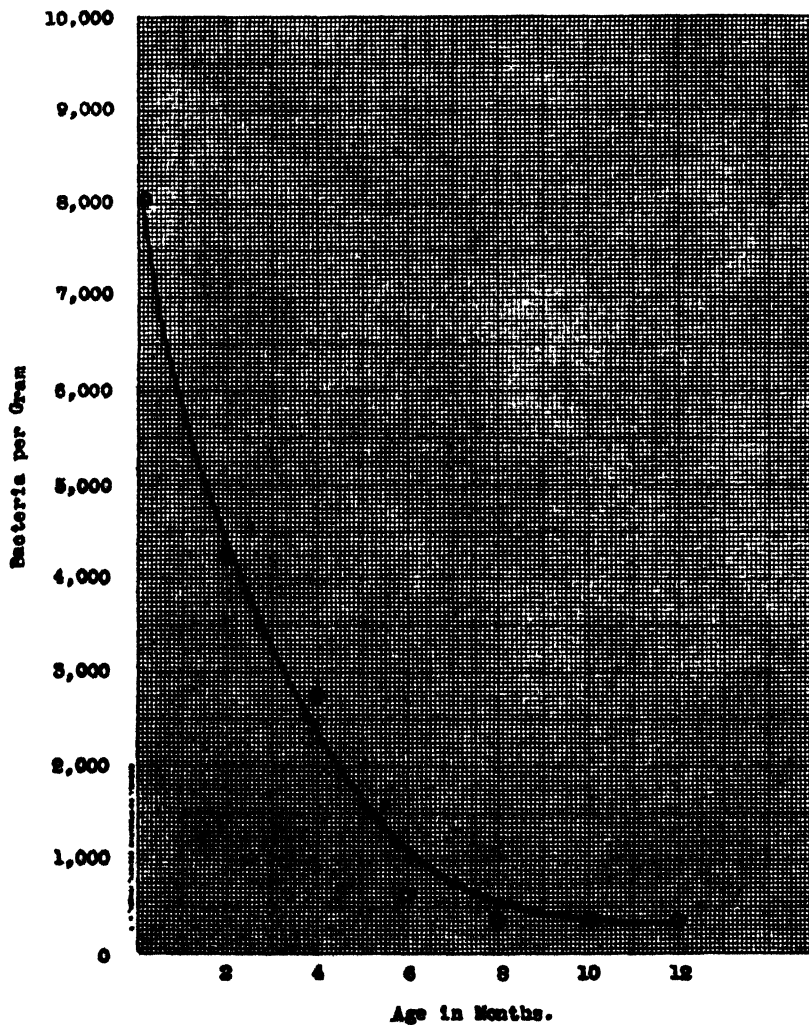


CHART "C"
Death-rate of Bacteria in Milk Powder
Average Moisture 7.04%

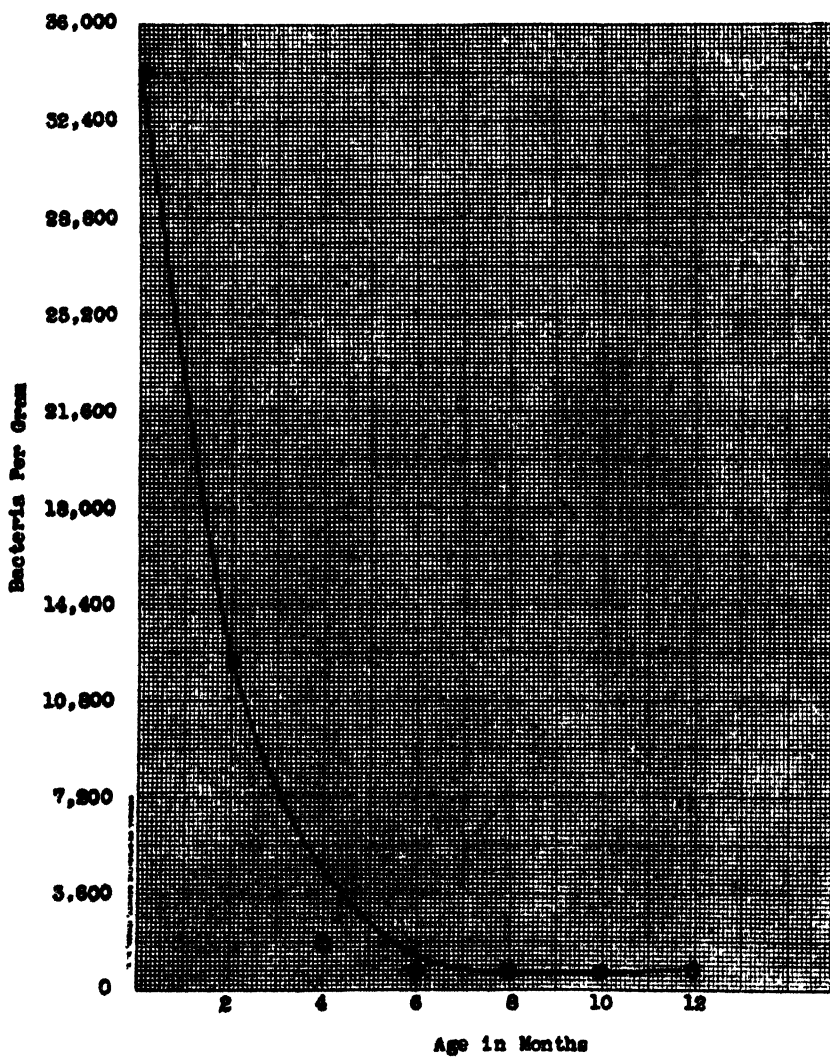
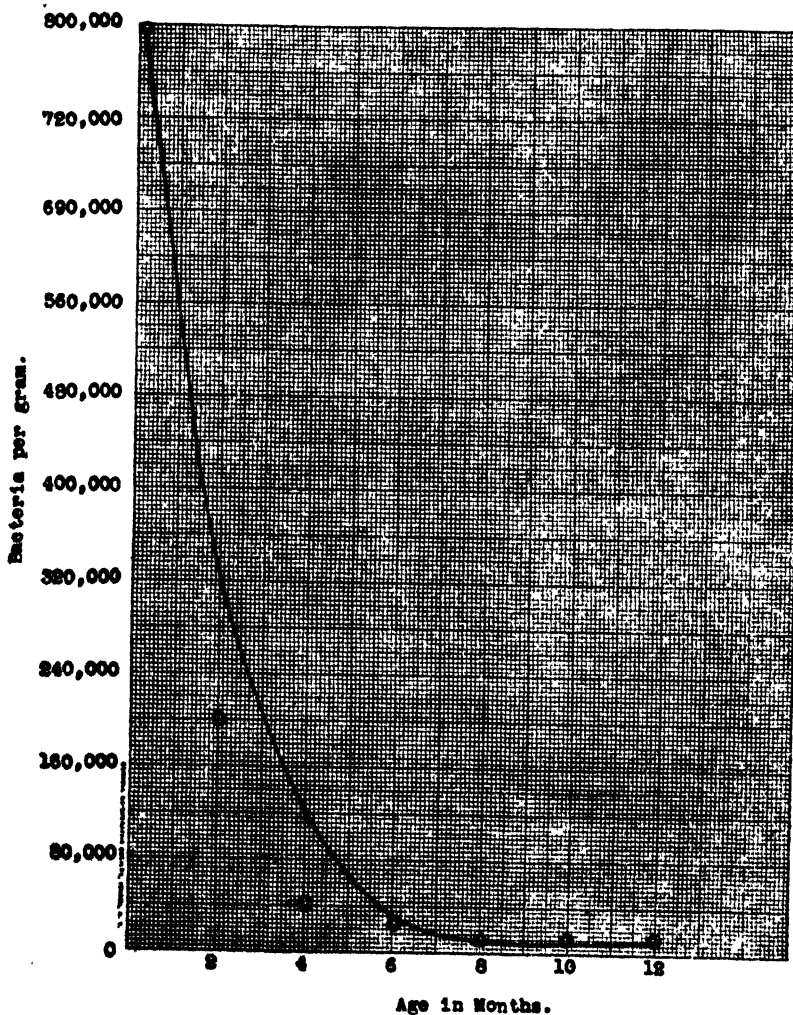


CHART "D"
Death-rate in Bacteria in Milk Powder
Artificially Inoculated.



CONCLUSIONS

From a review of the results recorded herein, the following conclusions may be drawn:

Bacteria over 1000 per gram in milk powder made by the Just process can be assumed in the majority of cases to be due to recontamination after the powder leaves the drying cylinders.

The bacterial content of the dried milk solids immediately after drying and before being subjected to recontamination did not seem to be affected by the number of bacteria in the liquid milk prior to drying providing such milk contains a normal flora.

The bacterial content of powder made by the Just process is normally lower than that reported in powder made by the spray processes.

The bacteria in milk powder die off rapidly during storage, and in normal powders made by the process under investigation reached a point of approximate constancy after two to four months.

The presence of large numbers of bacteria in desiccated milk does not produce any detectable effect upon the keeping quality in the presence of moisture concentrations which would admit the powder as a commercial article.

REFERENCES

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THE IMPROVEMENT OF QUALITY IN MILK THROUGH THE EFFORTS OF COLLEGE EXTENSION SERVICE

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To be most effective, agricultural extension service must solve the vital problems confronting the rural public. Methods leading to the solution of the various problems will necessarily vary because of different circumstances, but their ultimate effectiveness is measured by the extent to which agricultural practices are changed. Extension projects which yield results that can be measured and expressed in definite terms, carry greater weight than projects which do not lend themselves readily to measurement.

Certain types of extension projects are by nature more easily demonstrated than are others. Variety tests, and the influence of different fertilizers, are excellent illustrations of projects that yield results which may be tabulated or which may even be visible to the eye. This is, true only to a limited extent with reference to milk problems. It is easily possible to demonstrate the amounts of manufactured products made from milk of varying compositions, or to show the influence upon the temperature of different methods of cooling; but it is difficult to satisfactorily demonstrate such an abstract thing as quality in milk, except in research. Usually, however, such demonstrations are not practical nor workable from an extension point of view. The reason for this is largely due to the complications arising from the growth of microorganisms in milk, which introduces factors that are impossible to control under the conditions met in the field.

To mention the term "quality," as applied to milk, presupposed a definition of its meaning. Much space could be devoted to a discussion of the numerous factors involved, without even approximately settling the question. The factors that make up "quality" are complicated, and in general improvement in

quality has been retarded largely because of differences of opinion, making a common understanding impossible. While scientific circles have been debating as to what constitutes quality and as to means of controlling it, the consumer, the manufacturer, and the producer have been wrestling with the problem more or less aimlessly, because it was an immediate situation that had to be met in some way.

Just how the Extension Department could best help in bringing about actual improvement in quality, was a question difficult in itself to answer.

Since practically all changes in milk are the result of bacterial growth, it seemed that extension work which was designed to carry into the field knowledge concerning the relationship of bacteria to milk quality was the only really defensible procedure. Other factors that occasionally influence quality are not lost sight of; but the position is taken that the amount of milk which becomes undesirable because of these factors is relatively small, as compared to the amount that becomes undesirable through the accumulation of by-products resulting from bacterial action. If milk is produced and handled so as to keep the number of bacteria in it to a reasonable minimum, it must be agreed that there will, in general, be little reason for complaint against its quality.

The reducing of the number of bacteria necessarily involves the general question of bacterial "counts." The relative merits of different methods for making numerical estimates of bacteria will not be discussed here, further than to say that the differences between them are insignificant so far as the ultimate object—the improvement of milk quality—is concerned: nor will the use of bacteria "counts" as a means of expressing milk quality be discussed. There is much to be said on both sides of this question. The impelling reason for undertaking this project is the fact that thousands of dairymen in this state produce milk for fluid consumption in cities, and many of these cities have formulated ordinances enforcing numerical bacteriological standards.^{1, 2}

¹ The Sanitary Code established by the Public Health Council of the State of New York. Chapter III. Milk and Cream. New York State Department of Health, Albany, N. Y.

² Sanitary Code governing the production, pasteurisation, transportation handling, storage, sale and distribution of milk and its products. Department of Health, 505 Pearl St., New York City.

Although these standards have been in force for many years, the majority of dairymen are still in ignorance regarding bacterial relationships. This is no reflection upon their intelligence, because very little has been done to bring to them concrete information along this line. There is little reason to believe that much improvement in quality can be hoped for, until the dairymen appreciate the fact that microorganisms in milk are actual, biological entities, rather than products of imagination, and, are largely responsible for the undesirable changes; and until they learn the common sources of contamination and the conditions that promote or retard bacterial growth. In fact, this is the general thought underlying the project which is outlined below and which is so organized, it is believed, that data collected can be summarized to show definite results.

County.....
Year.....

New York State College of Agriculture
Sub-Project No. 1

Department of Dairy Industry
Extension Project No. 16

LEADERS: J. D. Brew, W. E. Ayres

OBJECT: To instruct the producers of milk concerning the sources and growth of bacteria and their relationship to the quality of milk; to assist in locating at any plant the principal causes for inferior quality; and to assist the producers in fulfilling the bacterial requirements for the various grades and for premiums.

PROCEDURE: This sub-project is to be conducted at any fluid milk plant or at any plant where milk products are manufactured. The work consists in grading the milk microscopically, according to the number of bacteria in it. In general the country grades established by the New York City Board of Health will be followed. The grades are as follows:

G = Good—qualifies for pasteurization as Grade A so far as number of bacteria is concerned. Less than 100,000 per cc.

Satis. = Satisfactory—qualifies for pasteurization as Grade B so far as number of bacteria is concerned. Less than 300,000 per cc.

Poor = Milk having a count greater than 300,000 per cc.

The milk delivered by each patron should be graded on at least two consecutive days and the results presented to the patrons at a meeting which has been previously scheduled, together with a discussion of the sources of bacteria and their growth. In order to ascertain whether or not any improvement in the quality of the milk resulted, it is necessary to regrade after the meeting. This may be done immediately following the meeting, or, if more convenient, at some later date. If the regrading is done immediately after, it is advisable to spend four days at the plant, especially if much of the milk is Poor in quality. If the quality is uniformly Good, it is unnecessary to

spend more than two days, or three at the most, in the community. A good idea is to plan work at two plants during the same week, and the entire project will be more satisfactory if, for instance, the milk at plant A is graded on Monday and Tuesday, with a meeting of the patrons on Tuesday evening, and the milk at plant B on Wednesday and Thursday, with a meeting on Thursday evening. Then return to plant A to regrade and check results on Friday, and to plant B on Saturday. This gives the dairymen at each plant an extra day in which to carry out the recommendations.

Personal visits to those farms from which inferior milk is being delivered will accomplish much in some cases.

COÖPERATION: The county agent will select the plants and make all necessary arrangements. There should be a real need for the work, which should be thoroughly appreciated by all parties concerned. The coöperation and interest of the patrons and the plant managers is essential. It is imperative that the patrons be acquainted beforehand with the purpose of the work, in order that they do not gain the impression that it is a mere inspection. To regard it as an inspection is a source of embarrassment and makes the benefits derived questionable. It is not necessary, however, to advertise the exact days in which the work is to be done.

The county agent should furnish the Extension Department with information concerning the following points: Type and name of plant, number of patrons, name of manager or superintendent, grade of milk handled, whether the grades are based on barn score or bacteria counts, whether or not premiums are paid, if a fluid milk plant the city in which the milk is sold, and the time at which the first patrons arrive in the morning.

EXPENSES: All necessary apparatus will be furnished by the Department of Dairy Industry. Local transportation will be furnished by the county agent. Other traveling expenses will be shared between the College and the Farm Bureau on the zone system.

This work had its beginning early in March, 1921, at a grade B pasteurizing plant, located in St. Lawrence County, from which milk was being shipped to New York City. Notice had been served upon the owners of this plant by the City Board of Health, to the effect that, because of too high bacteria counts in the milk at the time of delivery in New York, the milk would be excluded from the city if conditions were not improved within a certain specified time. This threat of rejection caused the owners and the patrons no small concern, because, if carried out, it would mean a serious financial loss to all. No one can, in justice, question the right of any city to enforce reasonable measures to safeguard its milk supply. However, when such notices are served, the management of most milk plants are invariably confronted with an almost impossible situation due to the fact that no one

knows where the trouble lies. It was to locate the source of the trouble that the College was called upon for assistance by the patrons, through the County Farm Bureau Manager.

The milk delivered by each patron was graded microscopically (1) on three successive days (March 7, 8, and 9), according to the procedure outlined. Grading on successive days gives more exact information as to the patrons delivering inferior milk, and also gives an impression of thoroughness which does much to stimulate the interest of the patrons in the entire project. Furthermore, it enables one to observe the plant operations more closely, because it may be that the cause of the trouble will be found in faulty plant methods, rather than in the quality of the milk

TABLE 1

The results of preliminary grading previous to meeting of patrons

DATE	NUMBER OF SAMPLES GRADED EACH DAY	NUMBER OF SAMPLES RATING AS		
		Good*	Satisfactory*	Poor*
March 7	72	56	5	12
March 8	80	63	6	11
March 9	85	65	5	14

* The terms *Good*, *Satisfactory*, and *Poor* are used because, under the conditions in New York State, they seem to better express to the patrons the quality of milk as regards meeting the bacterial requirements for Grade *A* or *B*. There is no thought of advocating their use to displace any of the present systems for designating or naming grades.

delivered by the dairymen. At a meeting of the patrons, the results of the grading were placed on a blackboard, as shown in table 1. The sources, growth, and control of bacteria were taken up in discussion.

The meeting was attended by one hundred and thirty persons, although there were only eighty-two patrons. One striking point brought out was the fact that a relatively large amount of Good milk was being delivered daily by the same patrons, and, for the most part, the Poor milk also was delivered by the same patrons. The bacteria count in much of the latter was so high as to jeopardize the whole supply after mixing. The large number of patrons delivering Good milk made a profound impression

and left no argument for the deliverers of Poor milk. The patrons demanded that the supply be graded later, to ascertain whether or not any improvement had taken place. This was done on March 30 and 31, but care was taken this time not to inform the patrons of the exact date. The results are shown in table 2.

The extent of the improvement can best be shown by giving in table 3 the percentage of the total number of samples that rated as Good, Satisfactory, or Poor before and after the meeting.

TABLE 2
Results of grading after meeting of patrons

DATE	NUMBER OF SAMPLES GRADED EACH DAY	NUMBER OF SAMPLES RATING AS		
		Good	Satisfactory	Poor
March 30	89	79	5	5
March 31	88	77	5	6

TABLE 3
Percentage rating according to grade

DATE	TOTAL NUMBER OF SAMPLES IN EACH GRADING	PERCENTAGE OF SAMPLES RATING AS			NOTES
		Good	Satis- factory	Poor	
March 7, 8 and 9	237	77.6	6.8	15.8	Before meeting
March 30 and 31	177	88.1	5.6	6.2	After meeting

The patrons asked for another meeting, which was held on the thirty-first with an attendance of one hundred and forty. The question of bacterial relationships was further discussed, and the patrons took full advantage of the opportunity offered to talk over frankly many of their own individual problems and experiences and to ask questions aiming to clear up some of the doubts in their minds. This is a most valuable part of any meeting.

The second grading revealed another interesting bit of information that greatly impressed the patrons. This was the marked decrease in the number of bacteria in the mixed milk from the vat after efforts had been made to improve the care and handling on the farm. On March 8 and 9, the count on the mixed milk

each day was 260,000 and 430,000 per cubic centimeter, respectively, but on March 30 and 31, the count was 80,000 and 140,000 per cubic centimeter, respectively.

The results of this first attempt to find out why a given supply of milk is high in bacterial content, and to carry to the producer information regarding the bacteriology of milk, exceeded expectations, and there has since been a large demand for similar work elsewhere. Fifteen plants in all have been studied, and many interesting facts could be presented if there were space to summarize and discuss separately, the results obtained at each. Practically all types of fluid milk plants were represented, including grade A plants at which premiums were paid for all milk delivered with a bacteria count, averaging under 10,000 per cubic centimeter, and plants where there was no effective control over quality and no particular stimulus to produce a better grade of milk. The general conditions surrounding one plant were quite different from those of another, and consequently the results obtained would naturally vary. The work at several of these plants yielded results as satisfactory and as encouraging as those obtained at the first plant. At two plants, however, the results were somewhat disappointing, although this fact occasioned no surprise because the patrons evidenced no particular interest in milk quality. There was no special incentive.

The summarized results obtained from the work done at all of the plants, together with the percentage of the total number of samples that rated as Good, Satisfactory, or Poor, are given in table 4.

The summary in this table represents forty-six days of work. In all, 3243 samples were graded, of which 64.5 per cent rated Good, 10 per cent Satisfactory, and 25.5 per cent Poor. The 3243 samples were taken from milk delivered by 1104 patrons. The total number of contacts at all of the eighteen meetings was 1056, which, if compared to the number of patrons, is nearly 100 per cent. This is somewhat misleading because at each of three plants an extra meeting was held at the request of the patrons. The average attendance was 58.7, the largest being 140 and the smallest 8. The average number of patrons at each plant was 61.3, the largest being 196 and the smallest 31.

At ten of the fifteen plants, data were obtained to determine whether or not any improvement resulted after placing the situation frankly before the patrons. Wherever possible, it is advisable to do this. Occasionally, however, the circumstances will be such that nothing particular is to be gained; such, for instance, as lack of interest, or time, or, as at one plant, when the milk delivered rated 97 per cent Good and a regrading would very likely be merely a repetition of results. The summary of the ten plants is given in table 5.

TABLE 4
Rating of all samples taken at fifteen plants

TOTAL NUMBER OF SAMPLES GRADED	NUMBER OF TOTAL RATING AS		
	Good	Satisfactory	Poor
3,243	2,088	327	828
Per cent.....	64.5	10	25.5

TABLE 5
Grading of all samples at ten plants before and after meeting of patrons

TOTAL NUMBER OF SAMPLES*	NUMBER OF TOTAL RATING AS						NOTES
	Good		Satisfactory		Poor		
		<i>per cent</i>		<i>per cent</i>		<i>per cent</i>	
1,446	847	58.6	153	10.6	446	30.8	Before meeting
1,097	852	77.7	97	8.8	148	13.5	After meeting

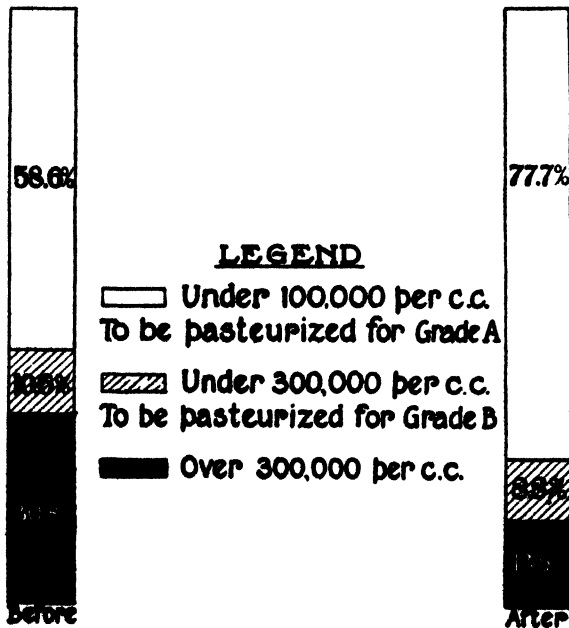
* Usually two days grading before meeting and one after.

The actual improvement may be shown more clearly in the accompanying graph.

Time, expense, and labor are important elements in conducting milk quality projects. The effectiveness of a project is very likely to be lost if either of these are unduly increased. It is highly desirable that all of the milk delivered be sampled and examined on the same day. To take part on one day and part on another, may yield results that are difficult to interpret because the conditions may not be comparable. Marked changes in outdoor temperature, for instance, influence the bacterial content.

The average fluid milk plant in New York State has from 50 to 150 patrons. With the direct microscopic method, the application of which will be described in a subsequent publication, it is possible in many plants for one man to take, prepare, and examine as many as 150 samples in one day. Ordinarily the examinations are completed early enough in the afternoon to make it possible to visit dairies from which Poor milk is being

AVERAGE QUALITY OF MILK AT 10 PLANTS BEFORE ^{AND} AFTER MEETINGS OF PATRONS



delivered and also to inform any patron if he wishes to know how his milk is graded. It occasionally happens that some patron would like to try a change in methods to see what the influence would be. For example, one patron placed two cans of night's milk in cold running water at 35° and two other cans were allowed to cool in the open air, which on this particular night was freezing point. On the following morning the count of the first two cans was 10,000 and of the last two, 900,000 bacteria per cubic centimeter

The temperature of the latter was 60° when delivered and that of the former 35°. This patron told at the meeting exactly what he had done, and there is no question that this in itself, did more to drive home the importance of prompt cooling than anything the speaker could have said.

There may be some justifiable doubts as to the advisability of making such experiments as this in the field. The writer at first proceeded with considerable misgivings, but several similiar experiences have been such as to demonstrate their value. On more than one occasion a certain point has been clearly shown in this way.

Absolute frankness is essential in presenting bacterial relationships to the dairymen. We have heretofore hesitated to discuss the question of bacteria counts frankly, for fear of misleading him, and instead have succeeded in doing what was hoped to be avoided. A dairyman wonders why his count "jumped from 40,000 to 50,000," and has been led to think that it was very likely due to some fault at the farm. The possibility and causes of variations should be frankly explained, and, instead of emphasizing insignificant differences in counts, the attention should be fixed on the fact that 75 or 80 per cent, or even more, of all the dairymen repeatedly produce milk good enough to qualify for grade A. This will do more to actually improve quality than anything else.

In organizing these projects, it is highly desirable, from the point of view of college extension, that every effort be made to eliminate all appearances of an ordinary inspection. The motive of an inspector is to catch some one off guard. This the dairyman resents. The motive underlying extension work with reference to milk problems is purely educational. The success of the project depends upon winning the confidence and the coöperation of every individual concerned. The object is to get at the facts leading to the elimination of all faults responsible for Poor milk, and thereby to ward off permanently the danger that a given milk supply will not meet the requirements of the Board of Health, or the possibility of the failure of the patrons to draw premiums or to meet other requirements related to quality.

To retain the confidence of all concerned (and that of the dairymen is particularly vital), the results obtained should never be used by an organization as a basis for rejection. It should be distinctly understood that any rejections following the project should be based upon data secured afterward by the organization itself. This plan can be quite satisfactorily carried out if all projects are organized, or at least sanctioned, by the County Farm Bureau.

In order to insure the most satisfactory results, it appears that some kind of stimulus is essential. Those dairymen who have the opportunity to draw premiums, or who sell to plants that are compelled to meet certain bacteriological requirements or where they feel that the bacteria counts made by the commercial company have not been fair, will be most interested in such projects conducted by the College. Absence of any stimulus whatever invariably means almost a total lack of interest.

In presenting and discussing the data contained in this paper, it is not assumed that the lowering of 30.8 per cent Poor to 13.5 per cent means that his improvement will necessarily be permanent. A 100 per cent perfection is practically impossible. The improvement of milk is primarily an educational project, plus some compelling force such as a financial stimulus or rejection of milk Poor in quality. Any change takes place slowly. The writer would not be surprised, for instance, to find at the place of the first project that the 6.2 per cent Poor on March 31 was somewhat higher, due to changes in patrons, or perhaps to a failure on the part of the plant management to enforce requirements, or to other factors. But there is good reason to believe that the general quality of this particular milk supply has been improved, because the procedure as outlined is an excellent way in which to carry to the patrons the message of the relationship between the bacteria content of milk and its quality.

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STRUCTURE OF POWDERED MILK AND ITS POSSIBLE RELATION TO THE KEEPING QUALITY OF WHOLE MILK POWDERS¹

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In the course of our investigations of certain chemical and physical properties of remade milks reported in the preceding paper, considerable difference was noted in the keeping quality of the whole milk powders used for reforming the fluid milks. The outstanding features of these observations were, first, that the predominating type of deterioration was a strong tallowy odor, and second, that this type of deterioration was confined to the spray process powders during the extent of the investigations (eleven months).

Coutts (1) has observed that whole milk powders deteriorate with age and that this is especially true of the whole milk powders made by the spray method. This investigator expressed the belief that oxidation of the fat is the cause of this deterioration. This harmonizes completely with the typical tallowy odor which we found developed in the spray powders, inasmuch as the tallowy decomposition of butter fat has been shown by Hunziker and Hosman (2) to be an oxidation reaction. It may be mentioned in this connection that the term rancidity is frequently applied to all types of fat deteriorations whereas it should, in the writer's opinion, be limited, in the case of butter

¹ Published with the approval of the Director as Paper No. 284, Journal Series, Minnesota Agricultural Experiment Station. The material presented in this paper forms a part of the thesis of C. D. Dahle, submitted in partial fulfillment of the requirements for the degree of Master of Science, in the Graduate School of the University of Minnesota, 1921.

fat, at least, to hydrolytic decompositions which result in the liberation of butyric and other volatile acids. Guthrie (3) has expressed this view also. Rancidity, in this sense, is not a common type of deterioration of whole milk powders, although it was noticed in a few cases with certain samples of centrifugal spray whole milk powder.

The fact that our samples of whole milk powders developed tallowiness on standing was confirmed in a chemical way by applying the test for "rancidity" worked out by Kreis (4). While this test is designated by Kreis as a test for rancidity, it is in reality a test for oxidation as Kerr (4) has pointed out. We applied this test to the fat secured from both centrifugal and pressure spray powders six to nine months old with positive results, the same test applied to fat from the drum process powders of the same age being negative.

No explanation has been advanced, so far as the authors are aware, of the difference in keeping quality of whole milk powders made by the spray and drum processes, which manifests itself in the production of tallowiness in the case of the former type of powder. Certain observations which the authors made on the structure of whole milk powders seem to offer a partial explanation, at least, for this difference.

It has been observed by others (1) that milk powders made by the spray process consist of microscopic granules which are spherical in shape. The authors found this to be true for both the pressure and centrifugal spray powders although the granules are considerably smaller in the former case than in the latter. Powder made by the drum process, however, consists of irregular platelets, as would be expected from the method of drying.

The significant feature of the structure of the spray process powders, however, is the fact that practically all the granules contain a spherical core of air, which does not appear at all in the case of the drum powders. This air cell is not discernible when the dry powder granules are observed under the microscope. However, if a drop of water is placed on the powder and the moistened powder observed, the protein material in the granules swells and becomes more or less transparent before disintegrating

and the core of air becomes plainly visible. At times the microscopic air bubbles will float around in the water like miniature balloons after the protein and fat have dissolved away. Again, the air cell will burst before the milk powder adhering to it has dissolved away, sending a shower of fat globules in all directions.

When one hinders the solution of the milk granule by chemical treatment, e.g., hardening in very dilute formalin, the swollen particle presents a very pretty picture showing the central spherical air cell surrounded by the dried serum solids in which are imbedded the globules of milk fat. If the fat globules have been stained with a fat dye² like Sudan III previous to the treatment with water, the picture presented is a very striking one. Figure 1 in the accompanying plate shows a photomicrograph³ of centrifugal spray powder after staining the fat globules and swelling the partially hardened particles. The core of air as well as the fat globules are plainly visible.

One noticeable difference between the centrifugal and pressure spray powders is in the relative size of the air cell in the granules. We observed that it usually forms a much smaller proportion of the total granule in the case of the pressure spray powder and was, at times, absent altogether. Several hours heating of this powder at 100°C. also caused the air to disappear. It was found more difficult to photograph the pressure spray powder granules to show the fat globules and the air cell because of the fact that the milk fat is homogenized in the process of manufacture, and also because it was harder to control the disintegration of the granules. Figure 2 shows a fairly successful attempt to obtain the picture of the disintegrating pressure spray powder granules showing the fat globules and some air cells. Figure 3 shows flakes of drum process powder after staining the fat globules.

² We found the simplest method of staining the fat was to dust a little powder into 60 per cent acetone and allow it to remain for a few minutes to partially coagulate the proteins. A few drops of N/50 HCl and a few drops of saturated acetone solution of Sudan III were added, the acid greatly increasing the intensity of staining. This mixture was allowed to stand a few minutes and the stained powder then washed by decantation using 20 per cent acetone.

³ Photomicrographs were made on Wratten and Wainwright's panchromatic "M" plates, using ray filters for contrast and detail.

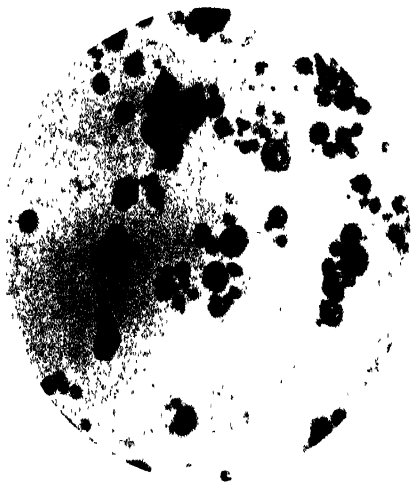


FIG. 1 Photomicrograph of hydrated centrifugal-spray whole milk powder granules, showing fat globules and central core of air. $D \times 170$. (Cut reduced one-third.)

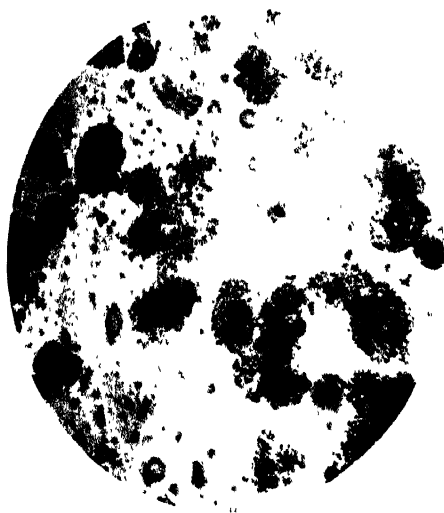


FIG. 2. Photomicrograph of hydrated pressure-spray whole milk powder granules, showing fat globules and central core of air. $D \times 330$. (Cut reduced one-third.)



FIG. 3. Photomicrograph of hydrated drum process milk powder, from partially skimmed milk, showing fat globules and absence of air from powder grains. $D \times 170$. (Cut reduced one-third.)

The low solubility of this powder in water does away with the necessity of hardening the flakes to prevent their rapid dissolution.

The tension which exists at the surface of the air cell and the surrounding membrane of proteins is surprisingly great. It was found that simple grinding in a mortar has no effect on it. Several hours' grinding in a ball mill was found necessary in order to break this tension and destroy the air cell.

We are unable to offer any adequate explanation for the interesting structure of the spray powders which we observe. Griebel (5) mentions having seen a milk powder full of air, his description resembling somewhat the one which is given in this paper. The cause of the air cell is evidently related to the fact that the milk is dried very rapidly in a large volume of air. It is possible, also, that a considerable amount of air is incorporated in the milk at some step in the process. The general picture of the dried particles, at least, suggests strongly the drying of a very fine foam.

The bearing of the structure of the spray process whole milk powders upon their inferior keeping quality, and their tendency to become tallowy, i.e., oxidize, is at once apparent. The fat is exposed not only to the air on the outside of the granules, which can be eliminated to a certain extent by packing, but also to air within the granules, which cannot be eliminated except by mechanical means or heat, both of which destroy certain desirable features of the powders. The fact that the pressure spray powders contain less air than the centrifugal spray powders is offset by the fact that the homogenized fat in the former presents a much greater surface for the action of the air. Whether the process of manufacture can be modified so as to prevent the incorporation of air within the granules, must be worked out in the factories.

It is well to bear in mind that the evidence that the structure of the milk powder particles has a bearing on their keeping quality is at present only circumstantial. We are engaged in submitting this hypothesis to experimental study and will report our findings in a later publication.

SUMMARY

The structure of whole milk powders made by the spray and drum methods is described. It is suggested that the presence of air within the granules of powder made by the spray process has an important bearing on the fact that this type of whole milk powder is especially prone to undergo tallowy, i.e., oxidative, deterioration.

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DAIRY NOTES

DAIRY DIVISION, B. A. I., UNITED STATES DEPARTMENT OF AGRICULTURE. Edwin H. Daane, a graduate of the University of Wisconsin, who specialized in dairy manufacturing, has been appointed for experimental cheese investigations, in the Dairy Division.

Henry B. Lowe, a graduate of Dartmouth College, who has taken graduate work at Columbia University, has been appointed for chemical investigations of dairy by-products (milk powder, etc.) Mr. Lowe has had several years' experience in commercial chemical plants before coming to the Department of Agriculture.

NEBRASKA UNIVERSITY. Mr. Benjamin Masurovsky, a graduate of the New Jersey Agricultural College, who has pursued graduate work at Columbia University and the University of California, joined the staff of the Dairy Husbandry Department of the University of Nebraska on January 1, 1922, as a graduate assistant. Mr. Masurovsky will assist in the research work of the department and will pursue his studies toward his doctorate degree.

WISCONSIN. At the October meeting of the Wisconsin branch of the Society of American Bacteriologists, Dean H. L. Russell was presented by his former students with a volume entitled "Papers on Bacteriology and Allied Subjects." This appreciated memorial was given in commemoration of the twenty-fifth anniversary of his doctorate. The real anniversary day occurred several years ago, but due to the war conditions immediately following, the publication of the volume was delayed.

It is a most comprehensive volume containing contributions from thirteen of the leading bacteriologists who were among the early students of Dr. Russell. E. G. Hastings of the University of Wisconsin reviews the Dean's scientific career and points out the strategic opportunities presented to pioneer bacteriologists. Dr. Russell was the first full-time agricultural bacteriologist in America. He was likewise one of the first men to be employed in this country to teach and do research work in bacteriology outside of the medical school. His scientific

papers, books, and bulletins, number well over one hundred and twenty-five and are of fundamental importance.

A development of the city milk supply problems is the contribution of H. A. Harding, formerly of the University of Illinois. He states the problems past and present in an interesting way and concludes by saying of Dr. Russell, "This Pioneer Bacteriologist in person and through his students has taken an honorable part in the solution of these problems."

That the greater prevalence of mold spores over bacteria in the air is due to the fact that most bacteria are readily killed by the sun's rays while mold spores are only slightly affected is the conclusion reached by John Weinzirl of Washington State University in his treatise on the resistance of mold spores to sunlight.

In a series of experiments carried on at the University of Minnesota, C. H. Eckles found that the percentage of fat in milk could be markedly increased for the first twenty to thirty days when it is followed by underfeeding during the period of lactation. Underfeeding of the cow must be taken into consideration in the interpretation of data involving variation in the composition of milk and butter fat.

L. A. Rogers, chief of the dairy division of the United States Department of Agriculture, summarizes the work done in his department on the characteristics of the Colon-Aerogenes group of bacteria. He regards *B. coli* and *B. aerogenes* as very distinct types. He discusses the taxonomic position of other members of this group in relation to these two varieties.

D. J. Davis of the medical school of the University of Illinois, presents evidence and argues convincingly to show that the fungus which causes sporotrichosis disease affecting both man and horses and common in France and occasionally in America, is identically the same species and should be called by the name first used by Hektoen in this country.

A butter having only a few yeasts and molds, when other conditions are favorable is a safer hazard for shipments and storage is the claim of F. W. Bouska and J. C. Brown of Chicago in their paper on "Yeasts and *Oidia* in Pasteurized Butter." Creameries which have the best commercial reputation for their butter also have the lowest yeast and mold counts. These two men give methods for sampling and counting butter which they have recently devised.

The late Dr. Edw. Birge presented his study on the activities of certain bacteria in sewage. He believed that some bacterial forms can

be found which will play an important rôle in the treatment of sewage, and that the time will come when septic tanks will be seeded as alfalfa fields and cream vats are seeded now.

A method for the detection of pasteurized milks is described in detail by Dr. W. D. Frost of the University of Wisconsin. The addition of a special dye stains the blood cells, always present in pasteurized milks. In raw milks the cells will not be stained.

A strong plea for the thorough investigation of all waters whose potability are questioned, and for thoroughly trained investigators experienced in laboratory and field work, is put forth by H. A. Whittaker of the University of Minnesota in a paper on the "Investigation of Drinking Water Supplies."

A. L. Amott, a commercial milk expert in Chicago has given much time, energy and thought to "The Milk Supply of Chicago," and discusses the source of supply, amount, production, transportation, city distribution, prices, farmers' organizations, and milk inspection. He calls attention to the improvement of the milk supply and the lowered baby death rate in recent years in Chicago.

B. W. Hammer of the Iowa Agricultural College in a paper on "The Bacteriology of Ice Cream" summarizes the knowledge of such points as number and kinds of bacteria, sources of materials, effect on the bacteria during freezing, hardening and holding, softening and rehardening. He also treats of the manufacture of ice cream with a low bacterial count, and the relation of ice cream to the public health, and bacterial standards.

J. H. F.

THE IDENTIFICATION OF THE BOVINE BY MEANS OF NOSE-PRINTS¹

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The various breed associations have always been confronted with a serious problem in the proper identification of animals for registration and of animals on official test. To guard against the possibility of substitution, the broken colored breeds (Guernseys, Holsteins and Ayrshires) require that a sketch of the color markings accompany the application for registry and that the actual markings of the animal on official test be compared by the test supervisor with the sketch on the registration certificate. This has been fairly successful but sometimes, due to the lack of drawing ability on part of breeders, the sketches in case of difficult markings do not agree with the markings on the animal. This frequently causes trouble when animals are sold or are on official test.

With the solid colored breeds, or breeds where a large number of individuals are of solid color, the present means of identification is even less satisfactory. Of these the Brown Swiss, Red Poll and Milking Shorthorn breed associations require no artificial markings while the Jerseys² require that each animal on test bear a tattoo mark in the ear. It is self evident that some natural distinguishing mark is necessary for positive identification as artificial marks, no matter how permanent they may be, can be applied in duplicate to different animals with which substitution can be practiced.

Under the present system for the identification of cows on official test, the word of the supervisor must be taken that he

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² The Jerseys also require the color of body, tongue and switch but according to T. J. Hooper of Kentucky, 66 per cent of Jerseys answer to the description: solid color, black tongue and black switch, so that such description does not positively identify.

has (in case of the broken colored animals) compared the color markings of the animal on test with the sketch on the registration certificate, and in case of the Jerseys, that he has examined the tattoo in the ear. In case of the other solid colored breeds, the owners word as to the identity of the animal must be taken.

It is with this problem in view that study was made of the practicability of the nose-print as means of identification. The work was started in October, 1921, from a suggestion of O. H. Baker of the American Jersey Cattle Club that the nose pattern might solve the identification problem if a satisfactory means could be devised of transferring the same to paper.

NO TWO ANIMALS HAVE BEEN FOUND WITH THE SAME DESIGN

Since October the nose-prints of more than 350 animals have been taken and carefully studied. No two have been found to be alike, or near enough alike but what they could easily be identified as being prints from different animals. For the purpose of study the prints were grouped into different types which are now being given further study. The basis for this grouping is the design formed by the lines starting from the center of the nose. According to this system, six distinct types have been worked out of which three are represented by figures 1, 3 and 5.

METHOD OF TAKING NOSE-PRINTS

The taking of the nose-prints has been found to be simple and easy. One man can easily take the print, when the animal is in a stanchion, by holding its head under one arm and taking the print with the free hand. Due to the fact that the bovine perspires freely through the pores of the nose, it is necessary to wipe the nose dry before applying the ink. For this purpose flannel cloth is used. The ink is then quickly applied by means of a stamping pad by either rubbing the pad back and forth or pressing directly against the nose. The print is then taken on the paper attached to a small board by pressing firmly against

the inked nose, beginning with the lower edge of the paper at the base of the upper lip and rolling toward the face.

In order to get clear prints the ink must be applied and the print taken quickly after the nose has been wiped dry as the moisture comes out rapidly from the pores. This causes the ink to run, filling up the grooves of the nose, and produces a smeared print.

The design is formed by the subcutaneous facial-nacial glands causing more or less pronounced elevations forming the irregular lines in form of grooves between these elevations. On some animals the elevations are less pronounced than on others, in which case the nose may be termed comparatively smooth. With "smooth" noses care must be taken not to press the ink pad too hard against the nose as such will fill up the groove with ink and the print will be smeared.

MIMEOGRAPH NEWS-PRINT PAPER GIVES BEST RESULTS

In tests with various papers mimeograph paper and news-print gave the best results due to their superior absorbing qualities. Trials with smooth finish papers proved that they are unsatisfactory, producing smeared or badly blurred prints. Yellow copy paper takes the ink very well but the pigment of the paper distracts from the cleancutness of the print, making it particularly unsatisfactory for photographic work.

Ordinary letterhead size paper cut into sheets $8\frac{1}{2}$ inches by $5\frac{1}{2}$ inches was found to be a convenient size to use. This gives ample room for two prints side by side and space for the necessary data at the top which consists of name of cow, name of owner and date. The blanks in use were made by a mimeograph, printing one set for each nose print to be taken or four on one letterhead size sheet.

A convenient way of handling the print paper is to attach, by means of a clip, a dozen or more of the above described blanks to a board $\frac{1}{4}$ inch thick and $8\frac{1}{2}$ inches long by $5\frac{1}{2}$ inches wide. After one set of prints is taken the sheet is torn off and a clean sheet exposed for the next print.

TESTS WITH INKS

Various inks were tried of which black stamping pad ink proved the most satisfactory with black printers ink and mimeograph ink close seconds. The chief objection to the latter two is the difficulty of getting a sufficient amount of ink into the pad to last for a large enough number of animals. Ordinary writing inks did not prove satisfactory due to their tendencies to run after applied to the nose. Blue and red printer's and stamping pad inks did not offer the contrast on the paper that the black inks did, and are not satisfactory for photographic work.

METHOD OF IDENTIFYING PRINTS

It is not necessary to have a perfect print in order to be able to positively identify it as being the same or different from another print. It is only necessary to have the details clear on a portion of the print. The first move in comparing two prints is to ascertain whether or not they are of the same type. Figures 1 and 2 are easily identified as being of the same type, straight lines emanating on either side of a central and perpendicular line of the prints and going outward and upward at about a 45 degree angle forming elongate figures. To positively identify figures 1 and 2 as being prints of the same animal, a small area (encircled) in the same region on the two prints is selected for detailed comparison. It is easily seen that the details within the circles in figures 1 and 2 coincide and that therefore they are two prints of the same animal. In the same manner figures 3 and 4 can be identified as the same animal, likewise figures 5 and 6.

Figures 1 and 3 can also be used to study type of nose-prints. In contrast to the description of figure 1 above, the lower lines of figure 3 radiate from a central point at the base of the print and branch forming irregular figures while the top lines are more or less horizontal. Figures 5 and 6 be can identified as a third type with the lower lines radiating from a central point at the base of the print as in figures 3 and 4 but do not branch and form rather regular elongate figures instead of irregular figures.



FIG. 1. NOSE-PRINT OF JERSEY COW HERD NO. 124 OWNED BY UNIVERSITY OF MINNESOTA

Print taken October, 1921



FIG. 2. NOSE-PRINT OF JERSEY COW HERD NO. 124 OWNED BY UNIVERSITY OF MINNESOTA

Print taken November, 1921. Easily identified as being the same as figure 1



FIG. 3. NOSE-PRINT OF JERSEY COW HERD NO. 113 OWNED BY UNIVERSITY OF MINNESOTA

Print taken October, 1921



FIG. 4. NOSE-PRINT OF JERSEY COW HERD NO. 113 OWNED BY UNIVERSITY OF MINNESOTA

Print taken November, 1921. Easily identified as being the same as figure 3



FIG. 5. NOSE-PRINT OF JERSEY COW HERD NO. 107 OWNED BY UNIVERSITY OF MINNESOTA

Print taken October, 1921



FIG. 6. NOSE-PRINT OF JERSEY COW HERD NO. 107 OWNED BY UNIVERSITY OF MINNESOTA

Print taken November, 1921. Easily identified as being the same as figure 5.

These cuts illustrate means of identifying prints. Only small portion of each print need be taken into consideration as shown by circles.

DOES PATTERN CHANGE WITH THE AGE OF THE ANIMAL?

Unless the pattern remains the same throughout the life of the animal the nose-print would have little practical use. To ascertain whether or not there is any change in the pattern as the animal grows older, prints have been taken monthly, commencing in October, for five successive months of five calves in the University farm herd. These calves ranged in ages (in October) from seven weeks to twelve months; therefore the prints taken for this purpose cover a span of from seven weeks to seventeen months. A careful study of these prints reveals that there is an enlargement of the nose but no change of the type or pattern.

Prints taken monthly of a large number of older animals for five consecutive months show no changes in type or pattern. From these observations as well as from the well established fact that the pattern of the human finger-print remains the same from infancy throughout life, it is reasonable to believe that the nose pattern of the bovine likewise remains constant throughout life.

PRACTICABILITY OF THE NOSE-PRINT AS A MEANS OF IDENTIFYING COWS ON OFFICIAL TEST

To give the nose-print system a practical test in relation to its use in connection with official testing prints have been taken by different official test supervisors of cows on test in various Jersey herds. A print of each cow on test in these herds was forwarded attached to the official test report sent to the American Jersey Cattle Club. None of these supervisors had seen a print taken, all instructions as to the method of taking the prints being given them by letter. The fact that all prints sent in were good proved that the method of taking nose-prints is simple enough. Reports from O. H. Baker, chief of the register of merit department of the American Jersey Cattle Club state that the prints sent in so far were very satisfactory. Prints have now accompanied the official test reports of all Jerseys on test in the University farm herd for four successive months and in eight other Jersey herds for from one to three months.

The owner of a Jersey herd reported that the supervisor had interchanged the names of two cows in making his official report for December. The supervisor when questioned about this said that it might be possible as both cows answered the same description but that he was certain that the report was correct as to the nose-print attached. As this was the first month of test the reports together with the nose-prints were held until the January reports were sent in, also with nose-prints attached, when a comparison of the two prints showed that there had been an interchange of names in the December reports. Had not the nose-prints accompanied the reports it would have been necessary to send the supervisor back to positively identify these animals.

OTHER POSSIBLE USES

In case of death of a solid colored, registered animal there is no means of guarding against the substitution of another solid colored animal in its place. If a nose-print accompanied the application for registration and that print is affixed to the registration certificate then substitution cannot be practiced without being detected.

It also has possibilities in connection with the writing of livestock insurance. Livestock insurance companies report that they are at times called upon to pay claims where they suspect that a dead animal which was not insured had been substituted for another one in the same herd answering the same description and which was insured. The nose-print will afford a means of positive identification in such cases.

SUMMARY

More work must be done on some phases of the nose-print as a means of identification. The work done so far may be summarized as follows:

1. No two animals have identical pattern and therefore a nose-print will enable positive identification.
2. The taking of nose-prints is simple enough to be practical.

3. It is possible to identify prints as being of the same or different animal even if they are not perfect.

4. The pattern remains the same throughout life.

5. It is practical for the identification of cows on official test and may prove valuable in connection with the registration of all solid color cattle.

6. It affords a positive means of identification when claim for loss is made under livestock insurance policies.

IS A PRELIMINARY DRY MILKING ESSENTIAL IN SEMI-OFFICIAL TESTS?

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The Kansas Agricultural Experiment Station rules governing Advanced Registry and Register of Merit testing require a so-called "dry milking" preliminary to the two day test. This dry milking is required in the breed association rules for Holsteins (9) and Guernseys (2), and in most cases for the Ayrshires (6), but until recently was not required by the American Jersey Cattle Club rules (3).

Very little experimental evidence concerning the influence of the preliminary milking seems to be available. Woodward and North (12) found that leaving strippings in the udder apparently increased milk production. No check period was included in the experiment, however, upon which to base comparisons. Eleven cows were included, some being used twice, making a total of 19 individual tests. From one-fourth to one-eighth of the milk was left in the udder on the evening of the second day. The results are shown in table 1.

It will be seen that leaving milk in the udders appreciably increased the milk and fat on the following day. Part of this may have been due to the residual effect of the milk which remained in the udders.

Work by Anderson (5) showed a wide variation in the fat production of consecutive milkings.

The recently published work of Regan and Mead (11) indicated that leaving one-half the milk from a previous milking in the udder caused an increase in milk and fat production, and also caused a larger percentage of butterfat in the milk during the following two days. This investigation was conducted with 4 cows, and included 7 experimental periods.

Since the dairy cattle breed associations depend largely upon two-day official tests as a basis for computing fat production in the Advanced Registry, and also depend upon the official two-day milk yield (2, 4, 7, 10) when computing abnormal or irregular milk reports (by the graph system), it is important to investigate the factors which may influence the two-day production of milk, butterfat, and the percentage of butterfat in the milk.

TABLE 1

*Effect of leaving milk in the udder, on the production of milk and butterfat,
Nebraska Agricultural Experiment Station*

DAY	MILK	FAT	FAT
	<i>pounds</i>	<i>per cent</i>	<i>pounds</i>
Second	369.6	4.681	17.301
Third	419.3	4.963	20.809
Fourth	396.5	4.852	19.238
Fifth	395.7	5.112	20.230

EXPERIMENTAL METHODS

A number of factors that may influence two-day and seven-day tests are being investigated at the Kansas Station, but the present article deals with the influence of the preliminary milking only.

In this study, animals of the four dairy breeds in the College dairy herd were used. High producing and low producing cows from each breed were selected. These included 11 Holsteins, 9 Ayrshires, 8 Jerseys and 8 Guernseys, making a total of 40 trials with 36 different cows. The total of 40 trials on 36 individuals included observations on 792 milkings, 440 milkings being used to compute the effects of leaving strippings in the udders.

Practically all of the cows were on Advanced Registry test, and the reports of the normal periods were forwarded to the respective breed associations, as the official report for the month.

The junior authors were present at each milking and feeding time throughout the investigation, to maintain regularity in hours and amounts of feed. Records were kept of weights of all

milk produced. Weather conditions were noted each day, and special notation made of weather changes.

The investigation included 5 separate nine-day experiments or trials at different seasons of the year, as outlined in table 2.

Each nine-day experiment was divided into five periods, as follows:

1. Two-day observation. A dry milking was taken on the evening of the second day.

2. Two-day normal official test conducted according to the rules of the American Dairy Science Association (1) for the conduct of official tests.

TABLE 2

Experimental periods, number of milkings per day, and animals used in investigations to determine the effect of dry milking on milk and butterfat production

EXPERIMENT	TIME (INCLUSIVE)	NUMBER OF MILKINGS DAILY	BREED AND NUMBER OF ANIMALS
1	December 7 to 15, 1920	2	4 Holstein, 4 Jersey
2	April 12 to 20, 1921	3	4 Guernsey, 4 Ayrshire
3	August 15 to 23, 1921	2	7 Holstein
4	September 27 to October 5, 1921	2	8 Jerseys
5	November 27 to December 5, 1921	2	5 Guernsey, 4 Jersey

3. One-day interval, at the last milking of which one-fourth of the milk as nearly as could be estimated, was left in the udders, an equal amount being left in each quarter, as nearly as possible. The quantity was estimated by milking out an amount equal to three-quarters of the average of the four previous corresponding milkings. Milkings on this day were weighed but not sampled.

4. Abnormal period. Two-day official test conducted according to the rules of the American Dairy Science Association, except that the dry milking was omitted.

5. Two-day observation period, during which the milk was weighed but not sampled.

EXPERIMENTAL RESULTS

Each experiment is reported separately in tables 3 to 8, inclusive, followed by a summary (table 9, 10, and 11) in which the total production is tabulated according to period and breed. Some of the results are presented graphically in figures 1, 2, and 3.

TABLE 3

Effect of dry milking in first experiment. December 7 to 15, 1920

BREED AND NUMBER OF ANIMAL		PRELIM- INARY PERIOD	NORMAL PERIOD	FIFTH DAY	ABNORMAL PERIOD	OBSER- VATION PERIOD
Holstein 89..	Milk, pounds.....	30.6	29.4	13.4	33.7	31.8
	Fat, per cent.....		3.885		3.809	
	Fat, pounds.....		1.1422		1.2838	
Holstein 53 ..	Milk, pounds.....	33.1	34.4	15.6	33.8	32.1
	Fat, per cent.....		4.194		4.445	
	Fat, pounds.....		1.4429		1.5024	
Holstein 112..	Milk, pounds.....	28.5	25.9	11.1	27.4	27.7
	Fat, per cent.....		2.942		2.924	
	Fat, pounds.....		0.7621		0.8011	
Holstein 104..	Milk, pounds.....	44.5	44.3	19.8	46.4	45.2
	Fat, per cent.....		3.509		3.672	
	Fat, pounds.....		1.5547		1.7037	
Jersey 302.....	Milk, pounds.....	34.2	36.0	15.6	38.1	35.9
	Fat, per cent.....		6.261		6.523	
	Fat, pounds.....		2.2540		2.4854	
Jersey 304.....	Milk, pounds.....	24.4	25.6	11.3	24.0	22.1
	Fat, per cent.....		4.064		4.273	
	Fat, pounds.....		1.0403		1.0256	
Jersey 311.....	Milk, pounds.....	40.3	43.3	19.2	45.5	43.5
	Fat, per cent.....		5.467		5.695	
	Fat, pounds.....		2.3671		2.5913	
Jersey 316.....	Milk, pounds.....	19.8	19.9	9.1	21.2	19.1
	Fat, per cent.....		5.726		5.721	
	Fat, pounds.....		1.1395		1.2128	
Total.....	Milk, pounds.....	255.4	258.8		270.1	257.4
	Fat, per cent.....		4.522		4.667	
	Fat, pounds.....		11.7028		12.6061	

Cows milked twice daily.

Weather conditions had no apparent influence, except in the fourth period of the fifth experiment, and will not be discussed in detail. At this time, an extreme drop in temperature ac-

TABLE 4

Effect of dry milking in second experiment. April 12 to 20, 1921

BREED AND NUMBER OF ANIMAL		PRELIM- INARY PERIOD	NORMAL PERIOD	FIFTH DAY	ABNORMAL PERIOD	OBSER- VATION PERIOD
Guernsey 429..	Milk, pounds.....	29.7	29.1	14.0	29.9	28.3
	Fat, per cent.....		4.925		4.830	
	Fat, pounds.....		1.4332		1.4441	
Guernsey 426..	Milk, pounds.....	45.4	44.0	20.6	45.1	42.4
	Fat, per cent.....		4.542		4.600	
	Fat, pounds.....		1.9986		2.0748	
Guernsey 417..	Milk, pounds.....	81.1	81.4	37.7	79.1	76.2
	Fat, per cent.....		3.575		3.375	
	Fat, pounds.....		2.9104		2.6696	
Guernsey 416..	Milk, pounds.....	36.7	32.2	14.6	36.3	33.6
	Fat, per cent.....		5.794		5.652	
	Fat, pounds.....		1.8658		2.0517	
Ayrshire 232..	Milk, pounds.....	62.8	62.1	28.4	61.3	59.2
	Fat, per cent.....		4.309		3.861	
	Fat, pounds.....		2.6761		2.3666	
Ayrshire 209..	Milk, pounds.....	66.0	70.8	32.4	78.5	72.9
	Fat, per cent.....		3.668		4.010	
	Fat, pounds.....		2.5972		3.1480	
Ayrshire 207..	Milk, pounds.....	70.9	66.1	32.7	69.3	66.6
	Fat, per cent.....		3.161		3.402	
	Fat, pounds.....		2.0897		2.3574	
Ayrshire 212..	Milk, pounds.....	84.6	79.9	37.4	85.2	83.5
	Fat, per cent.....		3.127		3.535	
	Fat, pounds.....		2.4986		3.0117	
Total.....	Milk, pounds.....	477.2	465.6		484.7	462.7
	Fat, per cent.....		3.881		3.945	
	Fat, pounds.....		18.0696		19.1239	

Cows milked three times daily.

accompanied by a high wind, caused a shrink of 1.2 per cent in milk flow of the entire herd of 71 cows, including the 9 cows on experiment. This probably affected the experimental results as mentioned later.

TABLE 5
Effect of dry milking in third experiment. August 15 to 23, 1921

BREED AND NUMBER OF ANIMAL		PRELIM- INARY PERIOD	NORMAL PERIOD	FIFTH DAY	ABNORMAL PERIOD	OBSER- VATION PERIOD
Holstein 113.	Milk, pounds.....	24.3	26.7	11.1	29.7	24.8
	Fat, per cent.....		4.199		4.334	
	Fat, pounds.....		1.1212		1.2871	
Holstein 102.	Milk, pounds.....	74.7	74.2	31.8	69.8	72.9
	Fat, per cent.....		3.472		3.549	
	Fat, pounds.....		2.5759		2.4774	
Holstein 127.	Milk, pounds.....	18.7	18.8	8.1	18.7	17.9
	Fat, per cent.....		4.390		4.606	
	Fat, pounds.....		0.8252		0.8613	
Holstein 121.	Milk, pounds.....	46.0	44.3	19.9	43.1	44.2
	Fat, per cent.....		3.666		3.640	
	Fat, pounds.....		1.6239		1.5687	
Holstein 110.	Milk, pounds.....	38.8	38.6	17.1	40.3	37.0
	Fat, per cent.....		3.338		3.254	
	Fat, pounds.....		1.2884		1.3147	
Holstein 106.	Milk, pounds.....	32.7	32.7	15.6	35.7	32.7
	Fat, per cent.....		3.470		3.498	
	Fat, pounds.....		1.1347		1.2489	
Holstein 129.	Milk, pounds.....	61.1	62.7	27.0	63.8	56.6
	Fat, per cent.....		3.164		3.202	
	Fat, pounds.....		1.9837		2.0426	
Total.....	Milk, pounds.....	296.3	298.0		301.1	286.1
	Fat, per cent.....		3.541		3.587	
	Fat, pounds.....		10.5530		10.8007	

Cows milked twice daily.

A summary of the results (table 8) shows a total increase of 48.9 pounds of milk and approximately 3.6 pounds of butterfat for the two-day period, as a result of leaving milk in the udder

on the day preceding the abnormal period. The percentage of butterfat was increased from 4.035 to 4.139.

Expressed on a percentage basis, the average increase is 3.13 per cent in milk production, 2.58 per cent in percentage of butterfat, and 5.82 per cent in production of butterfat.

TABLE 6

Effect of dry milking in fourth experiment. September 27 to October 5, 1921

BREED AND NUMBER OF ANIMAL		PRELIM- INARY PERIOD	NORMAL PERIOD	FIFTH DAY	ABNORMAL PERIOD	OBSER- VATION PERIOD
Ayrshire 212.	Milk, pounds.....	20 7	19 8	9.1	18.4	18.4
	Fat, per cent.....		3.833		3.863	
	Fat, pounds.....		0 7590		0.7107	
Ayrshire 207...	Milk, pounds.....	16 4	15 6	6.8	16.9	14.7
	Fat, per cent.....		3.518		3.517	
	Fat, pounds.....		0.5488		0.5943	
Ayrshire 209..	Milk, pounds.....	32.9	33.1	14.2	35.5	33.6
	Fat, per cent.....		3.822		3.477	
	Fat, pounds.....		1.2651		1.2345	
Ayrshire 214..	Milk, pounds.....	54.5	53.6	21.8	52.3	47.9
	Fat, per cent.....		3.658		3.870	
	Fat, pounds.....		1.9600		2.0242	
Ayrshire 237..	Milk, pounds.....	34.1	34.7	15.9	41.8	41.7
	Fat, per cent.....		3.893		3.845	
	Fat, pounds.....		1.3508		1.6073	
Ayrshire 234..	Milk, pounds.....	41.0	36.5	16.7	44.7	43.4
	Fat, per cent.....		3.448		3.774	
	Fat, pounds.....		1.2585		1.6872	
Ayrshire 208..	Milk, pounds.....	43.4	40.7	18.1	42.0	38.4
	Fat, per cent.....		3.501		3.775	
	Fat, pounds.....		1.4248		1.5855	
Ayrshire 236..	Milk, pounds.....	49.1	48.2	21.2	46.3	42.7
	Fat, per cent.....		3.573		3.821	
	Fat, pounds.....		1.7221		1.7693	
Total.....	Milk, pounds.....	292.1	282.2		297.9	280.8
	Fat, per cent.....		3.646		3.764	
	Fat, pounds.....		10.2898		11.2130	

Cows milked twice daily.

TABLE 7

Effect of dry milking in fifth experiment. November 27 to December 5, 1921

BREED AND NUMBER OF ANIMAL		PRELIM- INARY PERIOD	NORMAL PERIOD	FIFTH DAY	ABNORMAL PERIOD	OBSER- VATION PERIOD
Guernsey 433..	Milk, pounds.....	29.3	30.6	14.2	31.6	32.9
	Fat, per cent.....		4.168		4.569	
	Fat, pounds.....		1.2754		1.4437	
Guernsey 408..	Milk, pounds.....	27.3	27.0	12.6	25.4	25.8
	Fat, per cent.....		4.284		4.265	
	Fat, pounds.....		1.1567		1.0834	
Guernsey 417..	Milk, pounds.....	31.3	31.3	14.6	32.0	29.2
	Fat, per cent.....		3.968		4.002	
	Fat, pounds.....		1.2422		1.2805	
Guernsey 414..	Milk, pounds.....	23.9	26.7	12.5	25.9	23.2
	Fat, per cent.....		5.122		5.217	
	Fat, pounds.....		1.3676		1.3611	
Guernsey 435..	Milk, pounds.....	25.4	26.0	11.2	24.9	24.8
	Fat, per cent.....		4.123		4.212	
	Fat, pounds.....		1.0720		1.0488	
Jersey 322.....	Milk, pounds.....	26.1	24.3	11.1	23.8	26.7
	Fat, per cent.....		5.702		6.078	
	Fat, pounds.....		1.3855		1.4466	
Jersey 323.....	Milk, pounds.....	32.0	28.7	13.6	31.1	29.6
	Fat, per cent.....		4.951		5.694	
	Fat, pounds.....		1.4209		1.7707	
Jersey 312.....	Milk, pounds.....	27.2	26.6	12.1	26.5	24.2
	Fat, per cent.....		5.655		5.637	
	Fat, pounds.....		1.5042		1.4940	
Jersey 321.....	Milk, pounds.....	22.2	22.3	9.8	22.0	21.0
	Fat, per cent.....		6.413		6.508	
	Fat, pounds.....		1.4300		1.4318	
Total.....	Milk, pounds.....	244.7	243.5		243.2	237.4
	Fat, per cent.....		4.868		5.082	
	Fat, pounds.....		11.8545		12.3606	

Cows milked twice daily.

TABLE 8

Summary of experiments to determine the effect of dry milking on yield of milk and butterfat

EXPERIMENT	NUM- BER OF TRIALS		PRELIM- INARY PERIOD	NORMAL PERIOD	ABNORMAL PERIOD	OBSER- VATION PERIOD
First.....	8	Milk, pounds.....	255 4	258 8	270.1	257.4
		Fat, per cent.....		4.522	4.667	
		Fat, pounds.....		11.7028	12.6061	
Second.....	8	Milk, pounds.....	477.2	465.6	484.7	462.7
		Fat, per cent.....		3.881	3.945	
		Fat, pounds.....		18.0696	19.1239	
Third.....	7	Milk, pounds.....	296 3	298 0	301.1	286.1
		Fat, per cent.....		3.541	3.587	
		Fat, pounds.....		10.5530	10.8007	
Fourth.....	8	Milk, pounds.....	292.1	282.2	297.9	280.8
		Fat, per cent.....		3.646	3.764	
		Fat, pounds.....		10.2898	11.2130	
Fifth.....	9	Milk, pounds.....	244.7	243.5	243.2	237.4
		Fat, per cent.....		4.868	5.082	
		Fat, pounds.....		11.8545	12.3606	
Total.. ..	40	Milk, pounds.....	1,565.7	1,548.1	1,597.0	1,524.4
		Fat, per cent.....		4.035	4.139	
		Fat, pounds.....		62.4697	66.1043	

TABLE 9

Number of cows in each experiment showing an increase or a decrease; amount and per cent of increase summarized by factors

	EXPERIMENT					
	First	Second	Third	Fourth	Fifth	Total
Number of cows showing an increase in three factors.....	3	4	3	2	3	15
Number of cows showing an increase in two factors.....	4	2	2	4	2	14
Number of cows showing an increase in one factor.....	1	0	1	2	2	6
Number of cows showing a decrease in three factors.....	0	2	1	0	2	5

Actual and percentage increase

FACTORS	ACTUAL INCREASE	PERCENTAGE INCREASE
Milk, pounds.....	48.9	3.13
Per cent of fat.....	0.104	2.58
Fat, pounds.....	3.634	5.82

TABLE 10

Summary by breeds of the effect of dry milking on the production of milk and butterfat

BREED	NUM- BER OF TRIALS		PRELIMI- NARY PERIOD	NORMAL PERIOD	ABNORMAL PERIOD	OBSER- VATION PERIOD
Ayrshire.....	12	Milk, pounds.....	576.4	561.1	592.5	563.0
		Fat, per cent.....		3.591	3.729	
		Fat, pounds.....		20.1514	22.0967	
Guernsey.....	9	Milk, pounds.....	330.1	328.3	330.2	316.4
		Fat, per cent.....		4.362	4.378	
		Fat, pounds.....		14.3219	14.4577	
Holstein.....	11	Milk, pounds.....	433.0	432.0	442.4	422.9
		Fat, per cent.....		3.577	3.637	
		Fat, pounds.....		15.4549	16.0917	
Jersey.....	8	Milk, pounds.....	226.2	226.7	232.2	222.1
		Fat, per cent.....		5.576	5.796	
		Fat, pounds.....		12.5415	13.4582	
Total.....	40	Milk, pounds.....	1,565.7	1,548.1	1,597.3	1,524.4
		Fat, per cent.....		4.035	4.139	
		Fat, pounds.....		62.4697	66.1043	

TABLE 11

Number of animals in each breed showing an increase or a decrease in each of the three factors

FACTORS		AYR- SHIRE	GUERN- SEY	HOL- STEIN	JERSEY	TOTAL
Milk.....	Increase.....	8	5	7	4	24
	Decrease.....	4	4	4	4	16
Per cent of fat..	Increase.....	8	5	7	6	26
	Decrease.....	4	4	4	2	14
Fat.....	Increase.....	9	5	9	6	29
	Decrease.....	3	4	2	2	11
Total.....	Increase.....	25	15	23	16	79
	Decrease.....	11	12	10	8	41

The average for each individual experiment shows an increase in the production of butterfat and percentage of butterfat, and also in the production of milk, except in the fifth experiment when the production of milk remained practically the same. Failure to record a gain in this particular case may have been due to the marked change in the weather previously mentioned which apparently caused a decrease of 1.2 per cent in milk production of the entire College herd.

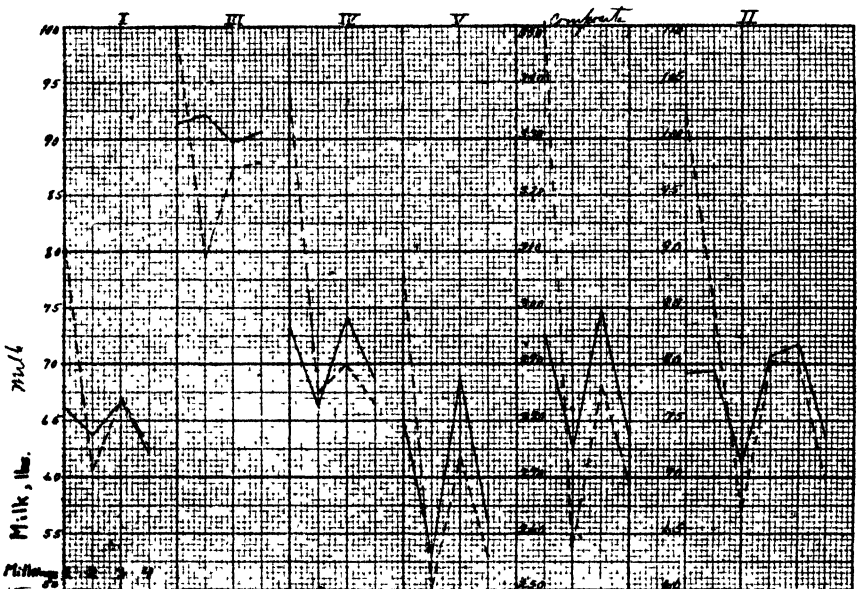


FIG. 1. EFFECT OF DRY MILKING ON MILK PRODUCTION
Normal period preceded by dry milking, ———.

Abnormal period; milk left in udder preceding this period, - - - - -

As indicated in figure 1, the greatest effect on milk yield appears to have been on the first and second milkings following the treatment. The effect on butterfat production extended over a longer period, and the percentage of butterfat over a still longer period. (Figs. 2 and 3.) This indicates that the effect on milk flow may be a residual effect, while the fat production is temporarily stimulated giving a greater percentage of fat in the milk.

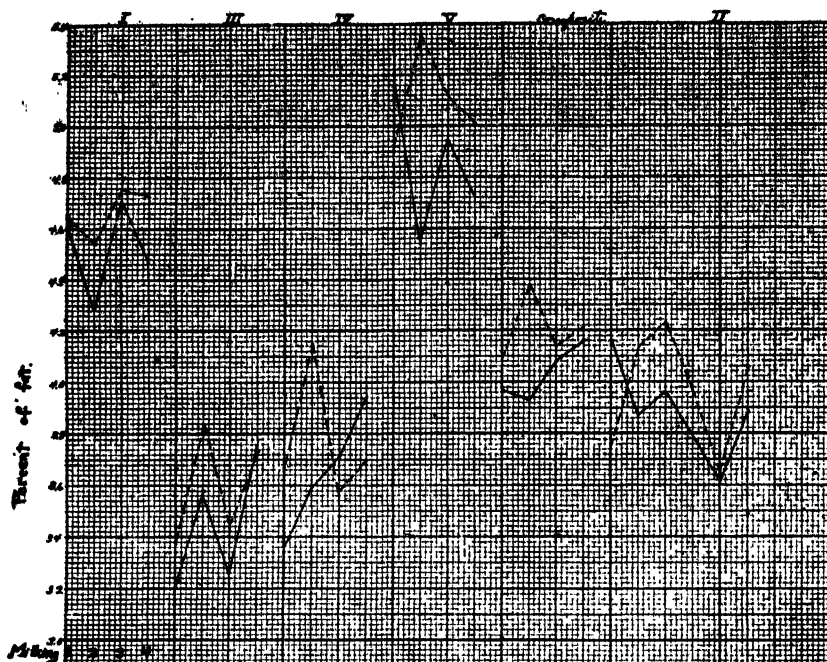


FIG. 2. EFFECT OF DRY MILKING UPON PERCENTAGE OF BUTTERFAT
 Normal period preceded by dry milking, ———.
 Abnormal period; milk left in udders preceding this period, - - - - -.

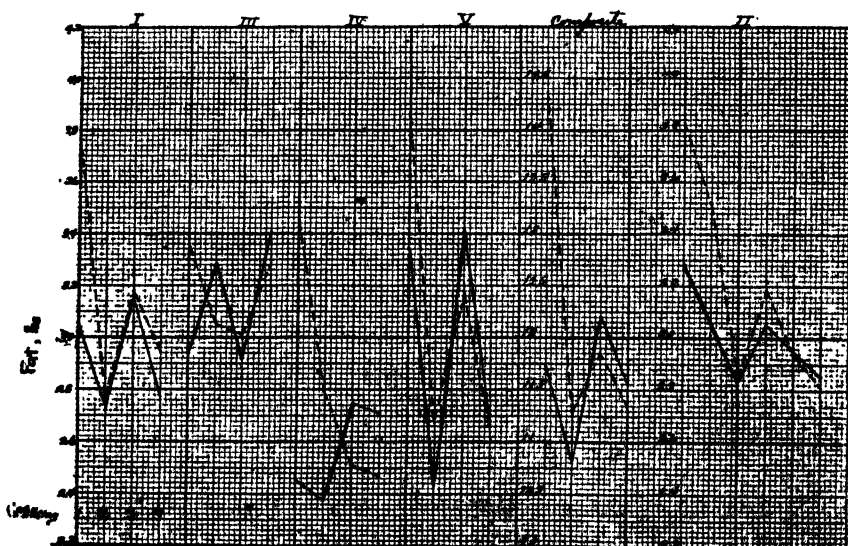


FIG. 3. EFFECT OF DRY MILKING UPON PRODUCTION OF BUTTERFAT
 Normal period preceded by dry milking, ———.
 Abnormal period; milk left in udder preceding this period, - - - - -.

Considering the data from another angle, it will be seen that leaving milk in the udder increased the test, and the production of milk and of butterfat in the case of 15 cows, while in the case of 5 cows there was a decrease in all three factors. Fourteen cows showed an increase in two factors, and 6 an increase in one factor only. This relation for each experimental period is shown in table 9.

When the data are assembled and studied (table 10), the factor of breed seems to have little influence. The ratio of increase to decrease (table 11) holds closely to 2:1 in all breeds, except with Guernseys where the ratio is 5:4.

It is of interest to determine what effect this general result has on the credit of a cow on semi-official test, where dry milking is not required. The example below, shows the effect on the credit of a cow producing 10,000 pounds of 4 per cent milk, when it has been necessary to chart by the graph method.

FACTORS	CREDIT	PERCENTAGE INCREASE	GRAPH RECORD
Milk, pounds.....	10,000	3.13	10,313
Per cent of fat.....	4.000	2.58	4.103
Fat, pounds.....	400.00	5.82	423.14

The credit in increased per cent of butterfat applies not only to the 10,000 pounds of milk actually produced by the cow, but also to that which is erroneously credited to her, thus having a greater effect upon the fat credit than on the milk credit.

It would seem, that if requiring a preliminary dry milking would eliminate the erroneous or dishonest credits possible to obtain through this practice, and thus semi-official records be rendered more reliable, the protection afforded to honest breeders would more than compensate for the extra expense of requiring a preliminary dry milking on all semi-official tests, with all breeds of cattle.

SUMMARY

1. It has been found possible, where a preliminary dry milking is omitted, and strippings left in the udder, to increase the yield

and percentage of butterfat in the milk during the following two days.

2. Total milk flow during the following two-day period, can be increased in this way, to conceal at least a 3 per cent padding of milk weights.

3. Breed as a factor, does not appreciably effect this influence.

4. The influence with high and low producing animals was relatively the same.

5. In view of these facts, a preliminary dry milking is essential, in order to obtain a representative "official" production during the two-day test in conducting semi-official records.

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THE CONDENSATION PROCESS OF PREPARING AN ICE CREAM MIX¹

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The general lack of uniformity in the use of materials and the methods of manufacture is responsible for the great variation in the standards and quality of ice cream found on the market. In the present method of manufacture of commercial ice cream, whole milk, sweet cream, plain (superheated) condensed milk, sugar, gelatin, and the desired flavoring usually form the ingredients. Evaporated milk, sweetened skim condensed milk, or milk powder and butter may be substituted for the above lacteal materials. A balanced supply of the different ingredients must be kept on hand at all times, and mixed together as needed. The mix is standardized to conform to a state requirement or a desired composition. In some cases the cream or the entire mix is pasteurized; and in a few plants the mix is homogenized. This must be followed by aging to insure the desired yield.

Within the last year the use of the condensation process of preparing an ice cream mix has greatly increased. This method consists essentially of combining sugar and the different lacteal substances in the proper proportions, evaporating to the desired composition, homogenizing, and cooling. The main purpose is to utilize merely milk and butter as the basis of the milk solids and butterfat. In the present methods of manufacture, however, it is impossible to use only milk and butter because the content of milk-solids-not-fat would be insufficient.

This paper is the report of a study which was made of the methods of preparation and standardization, and the bacterio-

¹ This work was carried on under the supervision of Dr. M. J. Prucha and Dr. O. R. Overman, and with the cooperation of Dr. H. A. Ruehe, Dairy Department, University of Illinois, Urbana, Illinois.

logical factors concerned in the condensation process. Part I deals with the preparation, and part II takes up a study of the bacteria in the condensed mix.

I. PREPARATION OF THE CONDENSED MIX

Apparatus required

In the preparation of the condensed mix the essential equipment is some form of concentrating apparatus. The ordinary "vacuum condensing pan" is the most efficient for this purpose. This consists of a copper retort with a steam-jacketed bottom, steam coils inside, and some form of condenser. A large, steam-driven, vacuum pump makes possible the evaporation of water under reduced pressure, at a temperature of about 130°F., thus promoting rapidity and economy of evaporation.

The ingredients are mixed in a large open kettle, commonly known as the "forewarmer." Any vat could be utilized for the purpose. Heat may be applied by means of coils, or by introducing live steam directly into the mixture. While the latter method introduces some water, it is more rapid and serves to agitate the milk while being heated.

A Baumé hydrometer, or a Westphal balance, may be used to indicate the point of proper concentration. Either a homogenizer or a viscolizer must be utilized in order to thoroughly emulsify the fat in the mix. This prevents the rise of butterfat during storage, and increases the viscosity of the mix. An efficient means of cooling must be provided. The ordinary upright open-faced cooler answers the purpose. This type of cooler also permits the drawing off of the mix directly into clean, dry cans. The mix may be pumped into large, insulated storage tanks and held until needed.

Materials used

Fresh whole milk is the best and cheapest source of the milk-solids-not-fat. This provides a clean, fresh-milk flavor which is not destroyed in the process. Economy is obtained by con-

tracting for milk directly from the producers. Even if milk is purchased from distributors, it is still the cheapest source of milk solids. Skim milk may be used to advantage, especially if an outlet for the sweet cream is available.

Unsalted butter is necessary to add to the mix in order to supply the proper amount of butterfat. Sweet cream may be used, but the use of it tends to increase the cost of the mix, unless there is no other outlet for the sweet cream. If sweet cream is used, advantage is taken of the solids-not-fat in the cream in the process of standardizing.

Either cane or beet sugar may be used for sweetening.

Process of manufacture

General procedure. 1. Examine the milk to make sure that it is fresh, clean, and sweet. Test a representative sample of the milk and of the cream (if cream is to be used) for percentage of butterfat and total solids, calculating the solids-not-fat by difference. The percentage of butterfat in the butter should also be determined.

2. Calculate the quantity of each ingredient to use:

a. It is necessary to first decide upon the percentage composition desired in the finished mix. Butterfat, milk-solids-not-fat, sugar, gelatin, and total solids must be taken into consideration.

b. Such amounts of the different ingredients are used as will give the desired proportion of butterfat, milk-solids-not-fat, sugar, and gelatin in the finished mix. No further standardizing will then be required unless the batch is overcondensed, in which case pure water only is needed.

c. When a definite quantity of mix is desired, the amount of milk required is ascertained by multiplying the percentage of milk-solids-not-fat in the mix by the pounds of mix, and dividing by the percentage of solids-not-fat in the milk. The amount of butter required is found by multiplying the percentage of fat in the mix by the pounds of mix desired, subtracting the product of the percentage of fat in the milk times the pounds of milk, and dividing by the percentage of fat in the butter.

d. When a given quantity of milk is used, the yield of mix is found by multiplying the percentage of solids-not-fat in the milk by the pounds of milk, and dividing by the percentage of milk-solids-not-fat in the desired mix. The butter is then calculated as in (c) above.

e. If cream is used, the ratio method of standardizing may be conveniently applied, advantage being taken of the solids-not-fat in the cream. Special tables have been prepared for a mix of a definite composition, which greatly simplify these calculations.

3. Weigh, or carefully measure, the required quantities of each ingredient into the forewarmer. The gelatin is added after condensing, either before homogenizing, or before freezing.

4. Heat the mixture in the forewarmer to 160°–170°F. This pasteurizes the mix, facilitates the melting of the butter and the solution of the sugar, and increases the rapidity of evaporation in the condensing apparatus.

5. Remove the excess water by evaporating under a vacuum of about 24 inches, at a temperature of 130°F. The batch is "struck" when the proper reading is obtained on the Baumé hydrometer, or Westphal balance.

6. Transfer the condensed mix to a vat with an efficient agitator. If this vat is mounted on scales, the mix may be weighed. Otherwise the full yield is obtained by measuring the volume and adding the necessary water to secure the proper yield.

7. Homogenize or viscolize the warm condensed mix, under a pressure of about 2000 pounds per square inch.

8. Cool the mix as rapidly as possible to a low temperature, preferably from 40° to 45°F.

9. Store the prepared mix at as low a temperature as possible, without freezing.

10. Retest the finished condensed mix, and restandardize if necessary.

Composition of a representative mix before and after condensing (100 gallons)

MATERIALS	TEST OF MIXTURE BEFORE CONDENSING	TEST OF THE CONDENSED MIX
pounds	per cent	per cent
Milk, 3.5 per cent	Butterfat..... 6.38	Butterfat.....10.0
fat; 8.7 per cent	Milk-solids-not-fat.. 7.66	Milk-solids-not-fat..12.0
solids-not-fat.... 1242.0	Sugar..... 7.66	Sugar.....12.0
Butter, 83 per cent	Gelatin..... 0.32	Gelatin..... 0.5
fat..... 56.0	Total solids.....22.02	Total solids.....34.5
Sugar..... 108.0		
Gelatin..... 4.5		
Total mixture.... 1410.5		
Water removed.....-510.5		
Finished condensed mix (100 gallons). 900.0		

The ingredients named under "materials" are placed in the forewarmer, resulting in a mixture having the composition shown in the second column. The excess water (510.5 pounds) is removed by condensing. When the desired composition is reached, the Baumé reading will be 10.3° at a temperature of 130°F. The final test is shown in the third column.

The mixture may be condensed slightly heavier than necessary, and made up to exactly 100 gallons by adding pure water. With the addition of flavoring, and gelatin if it has not been added previously, the mix is ready for freezing at any time.

Conclusions

1. A uniformly prepared ice cream mix may be made by the condensation process, using milk, butter or cream, sugar, and gelatin.

2. The condensed mix is easily standardized to a uniform composition by having the butterfat, milk-solids-not-fat, sugar, and gelatin in the proper proportion before condensing. A definite yield is thus obtained.

II. A BACTERIOLOGICAL STUDY OF THE CONDENSATION PROCESS

Since bacteria in ice cream are of importance from a sanitary as well as an economic point of view, a bacteriological study was made of the mix prepared by the condensation process.

Counts were made by the plate method, using standard lactose agar as media. An incubation period of four days at 30°C. was used. In all cases the counts given are the averages of the two plates made for each sample.

Source of bacteria

The lacteal substances used are the chief source of bacteria in the ice cream mix. Since milk and cream are ideal media for the growth and development of bacteria, it is to be expected that such products will greatly increase the bacterial count of the finished product. The sugar and gelatin are usually of minor importance, but utensils are a very prolific source of contamination.

The number of bacteria in the mix will vary according to the care that the milk products have received prior to their use, and according to the plant procedure. At the Iowa Experiment Station the number of bacteria in thirteen samples of ice cream was found to range from 130,000 to 40,850,000 per cubic centimeter (1). The United States Department of Agriculture found the average number of bacteria in 94 samples to be 37,859,907 per cubic centimeter. The maximum was 510,000,000, and the minimum 120,000 (2). The extremely high counts in ice cream are no doubt due to the fact that many manufacturers do not pasteurize their mix, nor take the proper precautions to avoid recontamination.

Efficiency of pasteurization of the condensing process

In order that ice cream may be a safe product for human consumption, and in order that the danger of souring may be reduced to a minimum, the mix should be subjected to some form of pasteurization. In the condensing process the mix is heated to 160°-170°F., in the forewarmer, and then condensed at a tem-

perature around 130°F. The efficiency of pasteurization of this process is shown by the following tabulation:

BATCH NUMBER	MIX BEFORE HEATING IN FOREWARMER	DIRECT FROM PAN		DESTRUCTION per cent
		Actual count	Comparative count*	
1	9,600,000	800	480	99.98
2	2,260,000	20,000	12,000	99.47
3	21,600,000	2,400	1,440	99.96
4	7,600,000	7,200	4,320	99.94
5	51,000,000	27,200	16,320	99.97
6	12,250,000	3,560	2,136	99.98
7	3,660,000	3,750	2,250	99.94

* The comparative count represents the number of bacteria per cubic centimeter that would be in the mix if the original volume had been retained, allowance being made for the decrease in volume, due to condensing. It is obtained by multiplying the actual count by 0.6.

These data show the completeness of pasteurization brought about by the heat in the forewarmer and condenser. In most cases the count of the condensed mix was quite low, the minimum being 800 and the maximum 27,200. Five of the seven batches run had a count of less than 7,500 per cubic centimeter.

The tendency of the unpasteurized mix to have extremely high counts is indicated by the number of bacteria per cubic centimeter found before heating it in the forewarmer. If ice cream is to be safely and economically produced, the mix must be subjected to some process that will greatly decrease the bacteria.

Number of bacteria at different stages of manufacture

To determine the relative importance of each step in the manufacture of the mix, counts were made of two batches after the completion of each step in the process. Care was taken to clean each utensil before it was used, by rinsing it with scalding water.

BEFORE HEATING IN FOREWARMER	DIRECT FROM PAN	DIRECT FROM HOMOGENIZER	AFTER ADDITION OF GELATIN	FROZEN ICE CREAM
9,600,000	800	1,400	1,450	2,600
2,260,000	20,000	26,200	26,250	31,000

These results indicate that after the mix is drawn from the pan the subsequent steps in the operation will result in the addition of only a very small number of bacteria if ordinary precautions are taken against recontamination. The increase after homogenizing and freezing is probably due, for the most part, to the breaking up of the bacterial clusters, which results in a higher count by the plate method.

The number of bacteria added by the gelatin will depend upon the grade of gelatin and the temperature to which it is heated. The temperature used in this experiment was 160° F., which was sufficient to kill most of the bacteria without injuring the jelling qualities of the gelatin. It may therefore be seen that with proper care there will be only a slight increase in the bacteria in the frozen ice cream over the number in the mix as it comes from the pan.

Keeping qualities of the mix

It often becomes necessary to store either the milk products or the mix for several days before freezing. It was found that condensed mix could be kept in a very good condition at a temperature of 32°-35°F. for a period of two weeks. Some was kept a month and then frozen into ice cream which contained no noticeable off-flavors.

Mix stored in 10-gallon can at 32°-35°F.

DAYS	BACTERIA PER CUBIC CENTIMETER	CONDITION
0	1,400	Very good
5	1,700	Very good
14	762,000	Good
23	42,210,000	Fair
32	188,500,000	Fair (frozen into ice cream)

As might be expected from the bacterial count, very little difference could be detected in the taste of the mix during the first two weeks of storage. It was not until the third week that a slight off-flavor began to develop. Thirty-two days after it was put into storage, the mix was frozen into ice cream, and al-

though it contained large numbers of bacteria, no off-flavor could be detected in the finished product. If the condensed mix is properly made and handled, the ice cream manufacturer should have no trouble in storing it for two weeks at a temperature of about 32° F.

Conclusions

1. From this experiment it may be concluded that it is possible to prepare an ice cream mix by the condensation process that contains a very small number of bacteria.

2. The number of bacteria in the mix as it comes from the pan approximately represents the number that will be in the frozen product, providing ordinary care is taken.

3. The mix will keep for two or three weeks when stored at a temperature of 32°-35°F.

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EFFECT OF STEAMING UPON THE GERM LIFE IN MILK CANS

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INTRODUCTION

For many years dairymen have commonly referred to milk cans which have been steamed as sterilized cans. It is, accordingly, something of a shock to learn that these steamed cans, as they reach the farm in summer, are usually teeming with germ life. Evidently there has been too much confidence in the killing effect of steam and this study is an attempt to measure the influence and limitations of steam in destroying germ life in milk cans.

For the past thirty years efforts at improving the city milk supply have been directed, largely, toward reducing its germ content and yet much of the milk as it reaches the milk plants during the summer months has a germ count of more than a million per cubic centimeter. The larger part of this germ life is the result of growth of bacteria in the milk. However, the morning's milk, which reaches the milk plant too quickly to show the influence of growth, in summer, commonly has a germ count of 50,000 to 100,000 per cubic centimeter. Previous studies (1) have shown that ordinarily at least 80 per cent of this germ life comes from the utensils in which the milk is handled, and among the utensils the cans in which the milk is transported from the farm to the milk plant are the outstanding source of bacteria.

Until recently it has been common practice to wash these cans at the milk plant, pass them over steam jets in an attempt to destroy the germ life in them, replace the covers and return them to the farm. In an increasing number of plants the cans, after being steamed, are exposed to jets of hot air with the object of drying

them. Except in rare instances this drying has not been complete, and as a result the interior of the cans as they are returned to the farm are commonly moist. At summer temperatures the presence of this moisture leads to a marked development of the germ life in the cans (2).

Data have already been presented (2) showing that when the cans containing this vast amount of germ life are properly rinsed with boiling water at the farm and used immediately, or are properly dried, they will add but few germs to the milk. However, it is manifestly undesirable that cans so heavily laden with germ life should be delivered to the producer.

Since in the milk plants steam is practically always applied by inverting the can over a jet of steam this method of application has been used in the present study.

OUTLINE OF THE STUDY

In the study there was determined the germ count of 1157 cans which had been steamed over a jet having a $\frac{1}{4}$ -inch opening. These cans were of 5-, 8-, and 10-gallon capacity and were used for shipping fresh milk from thirty-four farms to two dairies. Before being steamed, the cans were well washed in water containing about 1 per cent sodium carbonate washing powder.

In this study a simple form of steam jet was used. A strong galvanized iron plate 20 inches long and 12 inches wide was fastened horizontally at the side of the washing vat, and through the center of this plate the open end of the steam pipe protruded about 4 inches above the surface. The can to be steamed was inverted on this plate and the steam blown into it. The covers of the cans were steamed separately by placing them in a metal box and inverting the box over the steam jet.

The pressure at which the steam was blown into the can was measured by a sensitive steam gage placed about 18 inches from the jet opening, between the jet and the valve which admitted the steam.

The amount of steam which is blown into a can through a given jet during the process of steaming depends upon two factors;

the length of time of steaming and the steam pressure. Thirty-six different combinations of time and pressure were tested at each of which a number of cans were steamed and then examined for bacteria.

The examination for bacteria was made by rinsing each can with one liter (approximately one quart) of sterile water; after thoroughly shaking the can, a sample was taken of this water. The germ count of the sample was determined by the plate method, using standard lactose agar and incubating the plates five days at 20°C. and two days at 37°C. The germ count of the can was calculated from the germ count of the water. These results are expressed in terms of the germs per cubic centimeter which the can would add to the milk with which it was filled.

EXPERIMENTAL RESULTS

The amount of steam blown into a can at different pressures

The amount of steam blown into a can is controlled by three factors; the size of the jet opening, the length of time of steaming, and the steam pressure at the jet.

For the gage pressure of 15 pounds or more per square inch, the amount of steam flowing into the atmosphere may be calculated from the equation, $\frac{EP}{70} = \text{pounds of steam per second,}^1$ in which E = the area in square inches of the jet opening, P = the absolute pressure of the steam, and 70 is a constant.

For pressures below 15 pounds this formula does not hold; hence direct determinations were made by the following method: About 10 pounds of crushed ice was placed in a pail, and the steam was blown into it for a given time and at the desired pressure. The pail was weighed before and again after the steaming, and the difference in weight represented the amount of steam in pounds.

The amount of steam, at different pressures, which escapes per second when steam is blown into a can through a one-quarter inch opening is given in table 1.

¹ Kent. *Mechanical Engineer's Pocket Book*. 8th Edition, p. 844.

Bacteria in the cans before they were steamed

In making a study of the effect of any treatment one should of course be certain that the utensils used were sufficiently seeded with germ life to be typical examples. Since the process of determining the amount of germ life in cans removes a large portion of this germ life the information regarding the original germ content of the steamed cans was obtained by setting aside a representative portion of the washed cans to serve as checks. On each day, after the cans were washed and were ready to be steamed, a few cans were selected at random and were examined for bacteria without being steamed. In all, 191 such cans were thus examined. This was 14.2 per cent of all cans.

TABLE 1

Weight and volume of steam which escapes per second through an opening $\frac{1}{4}$ inch in diameter

STEAM PRESSURE	WEIGHT OF STEAM	VOLUME OF STEAM
<i>pounds</i>	<i>pounds</i>	<i>cubic feet*</i>
3	0.0064	0.1664
5	0.0104	0.2704
10	0.0159	0.4134
15	0.0199	0.5177
20	0.0242	0.6292
25	0.0280	0.7280
30	0.0315	0.8190
35	0.0350	0.9100
40	0.0385	1.0010
45	0.0420	1.0920
50	0.0452	1.1752

* The volume of the steam is computed from the weight by using 26 cubic feet as the volume of 1 pound of steam.

The examination of these cans showed that the effect of these cans upon the germ count of the milk, had it been poured into them, would have been very marked. If all the cans had been filled with milk, the average increase in the germ count of this milk, due to the condition of the cans, would have been 83,000 per cubic centimeter. This makes it evident that the cans which were steamed were abundantly supplied with germ life before being steamed.

TABLE 3

Relation between the amount of steam used and the bacterial count in the steamed cans

TIME STEAMED	PRESSURE OF STEAM	AMOUNT OF STEAM	AMOUNT OF STEAM	NUMBER OF CANS	CALCULATED CONTAMINATION OF MILK DUE TO:	
					Average can	Worst can
<i>seconds</i>	<i>pounds</i>	<i>pounds</i>	<i>cubic feet</i>		<i>germs per cc.</i>	<i>germs per cc.</i>
0	0	0	0	191	83, 186	1, 289, 000
3	10	0.048	1.248	21	1, 574	22, 048
5	5	0.052	1.352	12	44, 838	276, 667
5	10	0.079	2.054	24	386	2, 408
3	25	0.084	2.184	26	1, 166	12, 884
10	5	0.104	2.704	9	12, 080	45, 333
20	3	0.128	3.328	58	11, 923	184, 210
5	25	0.140	3.640	69	46, 784	1, 666, 667
15	5	0.156	4.056	10	5, 962	32, 667
10	10	0.159	4.134	65	22, 528	473, 684
5	35	0.175	4.550	5	246	789
30	3	0.192	4.992	55	3, 781	52, 368
20	5	0.208	5.408	23	603	3, 667
5	45	0.210	5.460	38	248	3, 777
5	50	0.226	5.876	36	100	1, 770
40	3	0.256	6.656	23	20	227
25	5	0.260	6.760	26	452	8, 000
10	25	0.280	7.280	29	20	447
30	5	0.312	8.112	36	269	4, 667
10	30	0.315	8.190	38	129	5, 789
20	10	0.318	8.268	22	799	933
50	3	0.320	8.320	30	159	2, 362
10	35	0.350	9.100	25	21	270
15	20	0.363	9.438	25	121	1, 867
35	5	0.364	9.464	70	1	34
60	3	0.384	9.984	35	37	449
25	10	0.398	10.348	28	24	237
20	15	0.399	10.374	24	11	138
15	25	0.420	10.920	39	8	215
30	10	0.477	12.402	73	2	45
20	20	0.484	12.584	58	6	135
25	15	0.498	12.948	51	7	152
30	20	0.726	18.876	50	2	14
30	25	0.840	21.840	6	1	1
60	25	1.680	43.680	6	1	1
120	25	3.360	87.360	6	1	1
180	25	5.040	131.040	6	1	1

Germ count of the steamed cans

A summary of the results of the examination of the 1157 steamed cans is given in table 2. The data are arranged in groups² according to the length of steaming and the pressure of the steam.

The volume inclosed by an 8-gallon can is practically 1 cubic foot. When the volume of steam blown into a can was equal to 2 cubic feet, the destruction of germ life became apparent. When the volume of steam amounted to approximately 5 cubic feet, the average bacterial count of the can after steaming was such that if the can had been filled with milk the bacterial count of the milk would have been increased less than 1000 per cubic centimeter. When the volume of steam was increased to about 9 cubic feet, the corresponding effect of the can on the milk was reduced to less than 100 per cubic centimeter. When the volume of steam was increased to 11 cubic feet per can, the average contamination of the milk was reduced below 10 per cubic centimeter.

Practical limits to the application of steam

It is evident from the data in table 2 that it is not practical to obtain sterile cans by the application of flowing steam. This was not accomplished when steam at a jet pressure of 25 pounds was blown into the can for three minutes.

In considering the application of these results to milk plant practice it should be remembered that with a $\frac{1}{4}$ -inch opening a steam pressure of 20 pounds at the jet is practically the working limit. The use of this steam pressure for sufficient time to produce a satisfactory reduction of germ life results in the loss into the room of a considerable volume of steam. Steam is expensive and its loss in this way is not only uneconomical but this waste steam seriously interferes with the comfort of the workmen and the drying of the cans. If the pressure is increased above 20 pounds this loss is so great as to be impracticable.

² The detailed data from these observations will later appear in a Bulletin of the Illinois Agricultural Experiment Station.

In considering can steaming it should be remembered that the producer who brings his milk to the milk plant waits at the receiving platform until his cans are returned to him. The washing and steaming must be so handled as not to delay the receipt of the next load of milk. Where the cans are washed, steamed, and dried at the central plant there is usually an equal necessity for haste in making the cans ready for the outgoing trains. To accomplish the work in the time available it is necessary that the minimum amount of time be given to the process.

By referring to table 2 it will be seen that after steaming the cans for five seconds with 25 pounds pressure the average contamination resulting from the cans amounted to over 40,000 bacteria per cubic centimeter. Where the cans were exposed for ten seconds with a steam pressure at the jet of 30 pounds the average contamination was reduced to 130 per cubic centimeter and after fifteen seconds exposure with 20 pounds pressure the contamination was still slightly above 100 per cubic centimeter.

Evidently steaming for ten seconds at workable steam pressure will not give satisfactory results, while fifteen seconds is so long a steaming period as to delay appreciably the return of the cans.

In practical operations it is necessary to provide a margin of safety, and this margin should be above 25 per cent. Observations in many plants suggest that under present plant practice the steaming of cans is rarely continued more than five seconds, and much of it is done in a shorter time. Evidently the present treatment would have to be increased about fourfold if the contaminating influence of the cans upon the milk put into them were to be reduced with certainty below 100 germs per cubic centimeter.

The limiting factor in the speed of handling cans is the time required to wash each can. Observations of hand washing in various commercial plants suggest that cans may be washed in about five seconds. Where a single steam jet is used, as is common practice in such plants, this practically limits the steaming to a like period. It is evident that the period of steaming may be lengthened by passing the cans over a succession of steam jets without seriously delaying the handling of the cans.

It should be kept clearly in mind, however, that putting a can into satisfactory condition for immediate use by steaming is an entirely different matter from treating it so that it will be in satisfactory condition ten or twenty hours later. The data presented in earlier publications (2) show that if the steamed cans remain moist and at summer temperature the growth of germ life in the can will soon render it unfit for receiving milk, and that the possibility of holding steamed cans in satisfactory condition is closely connected with the drying of the cans. Germs do not grow on dry tin.

HOW SHOULD CANS BE HANDLED

The data given in table 2 show that after 0.45 pound of steam is applied to a can it will increase the germ count of the milk poured into it less than 100 per cubic centimeter. It has previously been shown (2) that rinsing a can with 2 quarts of boiling water will reduce the germ count to practically the same point. Calculations of the heat relations show that the condensation of 0.45 pound of steam liberates 110,000 calories while the fall of 56 degrees noted when the 2 quarts of boiling water (2) were added to a can liberates 106,000 calories. Remembering that the hot water remains in the can while the uncondensed steam is lost into the atmosphere it is seen that the application of similar amounts of heat by either method produces like results in destroying germ life. Accordingly the question of whether a can should be treated with steam or with hot water is essentially a question of convenience. Moreover, the successive steps in the proper handling of cans vary somewhat, depending upon whether the cans are being washed by hand or by machinery.

Where the cans are washed by hand it is difficult to thoroughly rinse them with boiling water. In such cases steaming them over a jet tends to destroy germ life and rinses the interior through condensation of the steam. The more thoroughly this steaming is done the larger the reduction of germ life, but the steaming over a $\frac{1}{4}$ -inch jet must be continued fifteen to twenty seconds to produce, with certainty, cans which on being filled with milk will add less than 100 per cubic centimeter to the germ count of

the milk. Moreover, the cans should be thoroughly dried if they are to be held ten to twenty hours before being used. If the attempt to steam the cans is carried so far as to fill the air of the wash room with steam the proper drying of the cans is rendered doubly difficult. The drying of the can cover is even more difficult than the drying of the can, but unless the cover is also dried the labor of drying the can is largely lost.

Cans may be handled somewhat differently by machinery. The final step in the washing process should be to thoroughly rinse the cans with boiling water. It has been customary to pass the cans next over a number of steam jets and then over air jets. Where this is done the steam tends to fill the compartment where the cans are being dried and seriously interferes with the drying process. As a result such cans are rarely dried. Recent improvements in can handling machinery provide for a thorough rinsing with boiling water and minimize or omit the steaming process. The steam which was formerly applied to the cans is utilized in heating the air used in drying the cans. Observations of the cans coming from such machines make it evident that it is possible to provide cans in satisfactory condition either for immediate use, or for return to the producer.

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NEW METHOD OF BALANCING RATIONS

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With the advancement of scientific research in the field of animal nutrition the conception of a balanced ration assumes a somewhat different form.

1. Considerable emphasis is placed on proportioning the feeding stuffs according to their physical, chemical, and biological properties which affect food metabolism in the animal's body.

a. A feed is bulky or compact, coarse or fine, heavy or light, as considered under the heading, physical properties.

b. Palatability, acidity, alkalinity, toxicity, digestibility, and chemical composition of a feed, are properties of a chemical nature.

c. According to the biological effects exhibited by certain feeds on the digestive system, we find such properties as laxativeness, neutrality, costiveness or the property of causing constipation. Also the influences of "unidentified food substances" upon animal metabolism duly belong to the category of biological properties of food stuffs.

2. All feeding stuffs can be classified as follows:

Ration	Class I Roughages	{ Group I. Of high protein content. Group II. Of medium protein content. Group III. Of low protein content.
	Class II Concentrates	{ Group I. Nitrogenous feeds. Group II. Bulky (intermediary) feeds. Group III. Carbonaceous feeds.

3. For feeding milk cows the following feeding standards are widely used in this country: the modified Wolff-Lehmann or Henry and Morrison; Armsby's, and Haecker's systems. A fusion of their merits resolves itself into a "United Feeding Standard"

which facilitates the securing of all essential requirements in proper amounts.

4. Special consideration, therefore, should be given to the relation of digestible carbohydrates plus (digestible fat \times 2.25) to the digestible true protein set forth in such a feeding standard. The resulting ratio to be called the true nutritive ratio (T. N. R.).

5. While applying the new method of balancing^{*} rations for milk cows the following essentials are to be borne in mind:

a. The ratio of concentrates to roughages in every day practice of feeding milk cows should be known.

b. In compounding a ration it is important to consider the relation of the carbonaceous feeds to the nitrogenous feeds in the grain mixture varying with the kind of roughage used.

c. It is helpful to know the required physical condition of the ration, so far as bulkiness is concerned.

d. An estimation of the True Nutritive Ratio (T. N. R.) according to the United Feeding Standard, is an essential guide in balancing the daily ration.

EXAMPLE

Assuming that we are asked to balance up a ration for a Jersey herd averaging 30 pounds of 4 per cent milk daily per 1000 pounds live weight. The feed sources are as follows: Mixed timothy and clover hay is available and 10 pounds of it is fed daily per 1000 pounds live weight. The silage supply is limited and only 25 pounds is fed. In addition, soaked beet-pulp is used at the rate of 2 pounds, dry, to a cow weighing 1000 pounds. Corn is raised on the farm, and a home supply of corn and cob meal is available. The cheapest sources of purchased protein are secured from cottonseed meal, linseed oil meal, and gluten feed. Wheat bran is always on hand, and dried brewer's grain can be purchased to furnish more bulk.

SOLUTION

Step I. Calculate the standard requirements and the true nutritive ratio (T. N. R.) for the case under consideration (see

appended table 1). According to the United Feeding Standard the total daily requirements are:

TABLE 1

DIGESTIBLE CARBOHYDRATES PLUS (DIGESTIBLE FAT \times 2.25)	DIGESTIBLE TRUE PROTEIN	TRUE NUTRITIVE RATIO
15.842	1.970	1:8

Step II. Calculate the composition of the roughage fed daily per 1000 pounds live weight and the true nutritive ratio of the roughage.

TABLE 2

FEEDING STUFFS	POUNDS	DIGESTIBLE CARBOHY- DRATES PLUS (DIGESTIBLE FAT \times 2.25)	DIGESTIBLE TRUE PROTEIN	TRUE NUTRITIVE RATIO
Timothy and clover.....	10	4.455	0.36	
Corn silage.....	25	4.135	0.150	
Beet pulp, dry.....	2	1.309	0.014	
Total.....	37 cont.	9.899	0.524	1:18.8

Step III. Calculate: (a) The true nutritive ratio of the grain mixture to be compounded.

	DIGESTIBLE CARBOHYDRATES PLUS (FAT \times 2.25)	DIGESTIBLE TRUE PROTEIN	TRUE NUTRITIVE RATIO
Standard requirements	15.842	1.970	
Roughage provides.....	9.899	0.524	
Balance.....	5.943	1.446	1:4.1

Answer: 1:4.1 is the T. N. R. of the grain mixture under consideration.

Step IV. Place the available feeds into groups in respect to bulkiness, digestible true protein content, and digestible carbonaceous content (see appended table 2).

Group I. Bulky feeds available in the given case are: Wheat bran and brewer's grain.

Group II. Linseed oil meal, cottonseed meal, and gluten feed are high in digestible true protein.

Group III. Corn and cob meal is high in the digestible carbonaceous content.

Step V. Assuming that the proportion of the individual feeds within each group is as follows: In group I two parts of wheat bran are taken for every part of dried brewer's grains: in group II two parts of gluten feed and one and one half parts of oil meal are taken for every part of cottonseed meal.

Calculate the composition of each group its average true nutritive ratio, and "true protein factor."¹

TABLE 3

GROUP	PARTS	FEEDING STUFFS	DIGESTIBLE CARBONA- CEOUS CONTENT	DIGESTIBLE TRUE PROTEIN	TRUE PRO- TEIN FACTOR	AVERAGE TRUE NUTRITIVE RATIO
I	2	Wheat bran.....	0.968	0.216	7.19	1:3.37
	1	Dried brewer's grain.....	0.442	0.202		
			1.410	0.418		
II	1	Bulky feeds.....	0.470	0.139	3.81	1:1.94
			or 47%	or 13.9%		
	1	C. S. meal (choice).....	0.412	0.354		
	1½	Oil meal (O. P.).....	0.705	0.427		
	2	Gluten feed.....	1.186	0.402		
III			2.303	1.183	17.54	1:12.63
	1	Nitrogenous feeds.....	0.511	0.262		
			or 51.5%	or 26.2%		
	1	Corn and cob meal.....	0.720	0.057		
	1	Carbonaceous feeds.....	0.720	0.057	17.54	1:12.63
			or 72%	or 5.7%		

Step VI-A. Balance the three groups of feed available so that the grain mixture will give you the desired true nutritive ratio and the amount of every feeding stuff within each group.

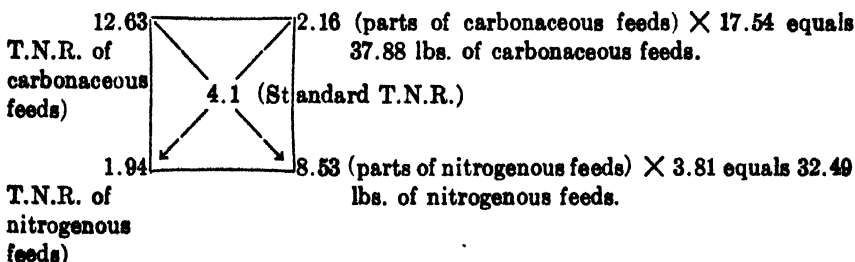
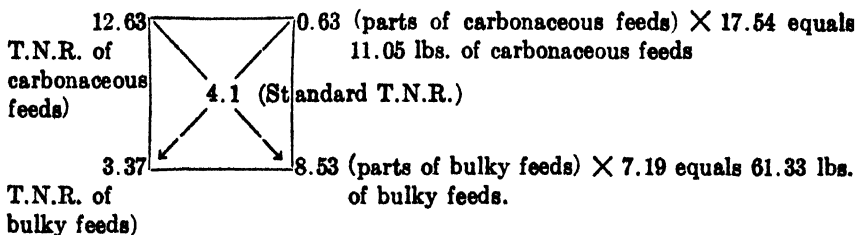
¹ The "true protein factor" is obtained by dividing 100 by the per cent of digestible true protein.

TABLE 4

	DIGESTIBLE CARBONACEOUS CONTENT	DIGESTIBLE TRUE PROTEIN	AVERAGE TRUE NUTRITIVE RATIO	TRUE PROTEIN FACTOR
	<i>per cent</i>	<i>per cent</i>		
Having bulky feeds containing.....	47.0	13.9	3.37	7.19
Having nitrogenous feeds containing.....	51.1	26.2	1.94	3.81
Having carbonaceous feeds containing.....	72.0	5.7	12.63	17.54

Apply Pearson's square method using the T. N. R. figures.

Thus:



Collecting the answers we obtain the following:

1. Carbonaceous feeds 11.05 plus 37.88 equals 48.93 pounds.
2. Bulky feeds, 61.33 pounds.
3. Nitrogenous feeds, 32.49 pounds.

The smaller number is always to be subtracted from the large one irrespective of position (provided you follow the arrow) in order to comply with the principle of alligation involved in the Pearson's square method.

Step VI-B. a. Since the bulky feeds are made up of three parts, namely, two parts of wheat bran and one part of dried brewer's grain.

Hence (1) $61.33 \div 3$ equals 20.44 pounds of dried brewer's grains.

(2) 20.44×2 equals 40.88 pounds of wheat bran.

b. In the case of the notrogenous feeds we have 1 plus 1.5 plus 2 equals 4.5 parts,

Hence: (1) $32.49 \div 4.5$ equals 7.22 pounds of c.s. meal (choice.)

(2) 7.22×1.5 equals 10.83 pounds of oil meal (O.P)

(3) 7.22×2 equals 13.44 pounds of gluten feed.

c. Finally the carbonaceous group of feeds is represented by 48.93 pounds of corn and cob meal.

TABLE 5

FEEDING STUFFS	AMOUNT	DIGESTIBLE CARBOHY- DRATES PLUS (DIGESTIBLE FAT \times 2.25)	DIGESTIBLE TRUE PROTEIN	TRUE NUTRITIVE RATIO
	<i>pounds</i>			
Corn and cob meal.....	48.93	35.229	2.789	
Gluten feed.....	14.44	8.562	2.902	
Oil meal (O. P.).....	10.83	5.165	3.086	
C. S. meal (choice).....	7.22	2.974	2.555	
Wheat bran.....	40.88	19.785	4.415	
Dried brewer's grain.....	20.44	9.034	4.128	
	142.74	80.749	19.875	1:4.06
Required T. N. R.				1:4.1

Step VII. By feeding 10.5 pounds of this grain mixture in addition to the amount of roughage allotted per 1000 pounds live-weight (see step II) a good supply of a 1:8 true nutritive ratio (T. N. R.) is insured.

I wish to express my thanks to Prof. H. P. Davis, Chairman of the Dairy Department and Prof. B. H. Thompson, Assistant Professor of Dairy Husbandry for reading the proof and for the valuable suggestions made in connection with this work.

APPENDIX

Merits of the main feeding standards used in this country

Henry and Morrison's feeding standard deals with the digestible crude protein and total digestible nutrients; Armsby's feeding standard is based upon digestible true protein and therms of net energy; and, Haecker's feeding standard considers the digestible carbohydrates and digestible fat, as well as, the digestible crude protein.

All of the enumerated feeding standards take into account the quality, as well as the quantity of the milk produced by the milk cow.

For each additional 0.1 of one per cent butterfat per pound of milk Henry and Morrison's standard requires from 0.0004 to 0.0008 pound of digestible crude protein and 0.006 pound of total digestible nutrients. Armsby's standard requires for each additional 0.1 of one per cent butterfat per pound of milk 0.0004 pound digestible *true protein* and 0.004 therm of energy. Haecker's standard assigns for each additional 0.1 of one per cent butterfat per pound of milk from 0 to 0.001 pound of digestible crude protein, from 0 to 0.01 pound of digestible carbohydrates, from 0 to 0.001 pound of digestible fat.

Illustration. Given a Jersey herd producing 30 pounds of 4 per cent milk daily per 1000 pounds of live weight. What are the daily requirements?

APPENDED TABLE 1

A 1000-pound cow producing 30 pounds of 4 per cent milk daily requires

FEEDING STANDARD	CASE	DIGESTIBLE CRUDE PROTEIN	DIGESTIBLE FAT	DIGESTIBLE CAR- BOHYDRATES	TOTAL DIGESTIBLE NUTRIENTS	DIGESTIBLE TRUE PROTEIN	NET ENERGY
			pounds	pounds	pounds	pounds	therms
Henry and Morrison's	Maintenance 1000 pounds, 30 pounds of 4 per cent milk	0.700			7.925		
		1.740 (av.)			10.380		
		1.53-1.95					
Total daily requirement.....		2.440			18.305		
Armsby's	Maintenance 1000 pounds, 30 pounds of 4 per cent milk					0.500	6.00
						1.470	8.10
Total daily requirement.....						1.970	14.10
Haecker's	Maintenance 1000 pounds, 30 pounds of 4 per cent milk	0.700	0.100	7.00			
		1.620	0.630	7.20			
Total daily requirement.....		2.320	0.730	14.20			
United feeding standard....	Maintenance 1000 pounds, 30 pounds of 4 per cent milk		0.100	7.00		0.500	
			0.630	7.20		1.470	
Total daily requirement.....			0.730	14.20		1.970	

$$\text{True nutritive ratio} = \frac{14.2 \text{ plus } (0.73 \times 2.25)}{1.97} = 8$$

$$\text{T. N. R.} = 1:8$$

APPENDED TABLE 2
Composition of some common feeds

NAME OF FEED*	DIGESTIBLE TRUE PROTEIN †	DIGESTIBLE CARBO- HYDRATES PLUS (FAT X 2.25) ‡	TRUE NUTRITIVE RATIO	BULK	REMARKS
	<i>per cent</i>				
<i>Concentrates:</i>					
Barley grain	8.3	70.4	8.4	Light	Neutral, fairly palatable
Buckwheat mid- dlings.....	20.8	52.0	2.5	Medium	Slightly constipative; poor for young stock
Corn meal.....	6.4	77.0	12.0	Heavy	Neutral; very palatable; widely used for feeding purposes
Corn and cob meal..	5.7	72.0	12.6	Medium	Neutral; fairly palatable
Cotton seed meal (choice).....	35.4	41.2	1.1	Heavy	Constipative; fair results in moderate feeding
Cocoanut meal.....	18.0	60.2	3.3	Heavy	Laxative; fairly palatable; can be used moderately
Cowpea meal.....	16.9	45.0	2.7	Heavy	Slightly laxative; good re- sults in all feeding pur- poses
Dried brewer's grains.....	20.2	44.2	2.1	Light	Neutral; fairly palatable, used extensively
Dried beet pulp	0.7	67.0	95.7	Light	Laxative; very palatable; excellent winter succu- lence
Flour middlings....	14.0	62.5	4.4	Heavy	Slightly constipative; poor for growing stock
Gluten feed	20.1	59.3	2.9	Medium	Neutral; fairly palatable; used for all feeding pur- poses
Gluten meal.....	28.1	53.8	1.9	Heavy	Neutral; fairly palatable; used for all feeding pur- poses
Ground oats.....	8.7	58.7	6.7	Light	Slightly laxative; very pala- table; excellent for all ing purposes
Hominy feed.....	6.5	77.2	11.8	Medium	Neutral; fairly palatable; used considerably
Linseed oil meal (O. P.).....	28.5	47.7	1.6	Heavy	Laxative; excellent for all feeding purposes

APPENDED TABLE 2—*Continued*

NAME OF FEED*	DIGESTIBLE TRUE PROTEIN †	DIGESTIBLE CARBO- HYDRATES PLUS (FAT × 2.25) ‡	TRUE NUTRITIVE RATIO	BULK	REMARKS
<i>Concentrates:</i>	<i>per cent</i>				
Linseed oil meal (N. P.).....	30.9	44.2	1.4	Heavy	
Malt sprouts.....	12.5	49.2	3.9	Light	Neutral; poor for growing stock
Peanut meal.....	16.9	38.5	2.2	Medium	Laxative; fairly palatable; good for all feeding purposes
Standard middlings	12.0	55.9	4.6	Medium	Slightly laxative; fairly palatable; good for all feeding purposes
Soy bean meal.....	27.3	45.1	1.7	Heavy	Laxative; good for all feeding purposes
Wheat bran	10.8	48.4	4.4	Light	Laxative; very palatable; excellent for all feeding purposes
<i>Roughages:</i>					
Alfalfa hay.....	7.1	41.0	5.7	Light	Laxative; very palatable; excellent nitrogenous roughage
Alfalfa green	1.9	11.3	5.9	Light	Laxative; very palatable; excellent roughage
Clover hay.....	4.9	43.3	8.8	Light	Laxative; very palatable; good nitrogenous roughage
Corn silage	0.6	15.8	26.3	Light	Laxative; very palatable; good for all feeding purposes
Millet hay.....	3.9	50.0	12.8	Light	Neutral; fairly palatable for ordinary purposes
Soy bean hay.....	8.8	41.9	4.7	Light	Neutral; fairly palatable; fair roughage for all feeding purposes
Timothy hay.....	2.2	45.5	20.6	Light	Constipative; poor dairy roughage
Mangels.....	0.1	8.0	80.0	Light	Laxative; very palatable; excellent for all feeding purposes

* Computation made from composition table of Pennsylvania Experiment Station Bulletin 142.

† Computed from Henry and Morrison, "Feeds and Feeding."

‡ Taken from Bulletin 90, Storrs Experiment Station.

STUDIES IN THE GROWTH AND NUTRITION OF DAIRY CALVES

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I. THE GESTATION PERIOD

The gestation period of the cow is generally recognized as of fairly definite length, though few reports have been made on this subject. These reports include a considerable number of animals but little study has been given to the variations which occur in the length of gestations.

RÉSUMÉ OF PREVIOUS WORK

On summarizing 764 gestation periods, Spencer (4) found that 314 of them ended 284 days or less after the date of breeding, and 310 of them 285 days or more after that time. He stated that gestations of less than 260 days' duration had ended decidedly prematurely, while those of over 300 days were very irregular though they did not affect the offspring. After studying 182 normal gestations on 20 cows, Wing (5) found the average length to be 280 days regardless of the sex of the offspring. The majority were from 274 to 287 days in length and were fairly evenly distributed throughout that period. He also noted that some cows had uniformly long and others uniformly short periods of gestation. From 1062 observations Fleming (2) found the average length of the gestation period to be 283 days.

EXPERIMENTAL WORK

The data presented here was collected from the herd on Iowa State College Dairy Farm from December, 1907, to November, 1921, inclusive. A summary of 369 gestation periods are presented. Gestation periods during which twins were carried have

not been included. In addition to the general results obtained an effort has been made to find what influence, if any, the age of the cow at the time of freshening and the season of freshening have on the length of the gestation period.

TABLE 1
Average length of gestation period

BREED	NUM- BER OF COWS	NUMBER OF CALVES			AVERAGE GESTATION PERIOD		
		Male	Female	Total	Male	Female	Total
Purebreds							
Ayrshire.....	14	15	20	35	days 277	days 279	days 278
Guernsey.....	25	42	35	77	282	281	281
Holstein.....	28	41	33	74	276	279	278
Jersey.....	31	38	35	73	280	280	280
Grades							
Ayrshire.....	2	2		2	282		282
Guernsey.....	21	22	26	48	282	282	282
Holstein.....	14	20	17	37	279	280	279
Jersey.....	10	7	12	19	279	277	278
Scrubs.....	4	2	2	4	287	285	286
Grades.....	47	51	55	106	280	280	280
Purebreds.....	98	136	123	259	279	280	279
All calves.....	149	189	180	369	279	280	280

TABLE 2
Distribution of gestation periods according to their duration

LENGTH OF GESTATION	NUMBER OF CALVES			DISTRIBUTION OF GESTATIONS		
	Male	Female	Total	Male	Female	Total
<i>days</i>				<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
251	15	2	17	8.4	1.1	4.7
261	23	9	32	12.7	5.2	9.1
271	69	80	149	38.3	45.7	42.0
281	69	75	144	38.3	42.8	40.6
291	4	9	13	2.3	5.2	3.6

The gestation periods have been tabulated by breeds, and the scrubs separated from the grades and purebreds. The scrubs

TABLE 3

Influence of age of cow at freshening on length of gestation period

AGE OF COW	NUMBER OF CALVES			AVERAGE LENGTH OF GESTATION		
	Male	Female	Total	Male	Female	Total
<i>years</i>				<i>days</i>	<i>days</i>	<i>days</i>
1	1	2	3	285	280	281
2	44	39	83	282	279	281
3	41	32	73	280	279	280
4	26	31	57	282	279	280
5	20	20	40	285	279	282
6	11	14	25	283	280	281
7	13	12	25	282	281	282
8	12	10	22	280	282	281
9	4	6	10	282	285	284
10	3	4	7	281	285	283
11	4	3	7	284	284	284
12	4		4	281		281
13	1	2	3	291	278	282
14	1	2	3	285	282	283

TABLE 4

Influence of season of freshening on length of gestation period

MONTH OF FRESHENING	NUMBER OF CALVES			AVERAGE LENGTH OF GESTATION		
	Male	Female	Total	Male	Female	Total
				<i>days</i>	<i>days</i>	<i>days</i>
January.....	13	19	32	281	281	281
February.....	20	23	43	278	280	279
March.....	26	22	48	280	280	280
April.....	15	11	26	279	284	281
May.....	11	14	25	279	278	278
June.....	14	10	24	278	282	280
July.....	15	11	26	277	285	280
August.....	11	14	25	283	280	282
September.....	14	16	30	278	279	279
October.....	18	19	37	281	284	284
November.....	17	9	26	277	283	279
December.....	14	8	22	284	281	283

and grades were all used for experimental breeding work. A sex distinction was also made in the tabulation.

In order that a clearer conception of the connection between the sex of the calf and the length of gestation period may be obtained, the percentage of the calves of each sex carried through gestation periods of various lengths have been determined. Only 355 gestations could be used for this purpose and they are tabulated in groups varying in duration by ten days. The first group includes gestations of 251 to 260 days, the second group is made up of gestations of 261 to 270 days, and so on. Gestations of less than 260 days and those approaching 300 days in length must be looked on as somewhat abnormal although the calves were dropped in good healthy condition.

In determining the influence of the age of the cow on the length of the gestation period, some of the periods had to be discarded as records were not available in all cases of both the age of the cow and the length of the gestation period. As a consequence only 362 of the 369 periods are used for this purpose.

In determining the influence of the season of freshening on the length of the gestation period, the records were tabulated by months. It was again necessary to delete a few records so that only 364 are available.

DISCUSSION OF RESULTS

When all the records available for this study are taken into consideration, it is found that the average length of the gestation period for dairy cows is 280 days. There were no significant variations so far as the breed of the cows was concerned.

It has frequently been stated and is a very common belief that the periods of gestation during which bull calves are carried are longer than those for heifer calves. The average of all the records studied here, however, shows an average gestation period of 280 days for both bull and heifer calves. On studying the distribution of the gestation periods of various lengths, it is found that 38.3 per cent of the male calves were dropped after a period of 271 to 280 days in utero while an equal percentage of them were dropped after 281 to 290 days in utero. The

corresponding figures for the heifers are 45.7 per cent and 42.8 per cent. This shows that the gestation periods are more closely grouped, so far as duration is concerned, in the case of heifer calves than in the case of bull calves. When the extremes are considered, it is found that 21.1 per cent of the bull and 6.3 per cent of the heifer calves are carried in utero for 270 days or less while 2.3 per cent of the bull and 5.2 per cent of the heifer calves are carried for 291 days or more. This does not indicate any greater length of the gestation period in the case of bull calves.

The age of the cow at the time of freshening and the season of freshening are factors which also appeared to have no influence on the length of the period of gestation.

SUMMARY

From the data studied here it was found that:

1. The average length of the gestation period in dairy cows was 280 days.
2. This was not affected by the breed of the cows.
3. The sex of the calf had no apparent influence on the length of the gestation period.
4. Male and female calves were dropped in practically equal numbers.
5. On the average 82.6 per cent of the calves were carried in utero for a period of 271 to 290 days.
6. The age of the cow at the time of freshening was without influence on the duration of the gestation period.
7. The length of the gestation period was not influenced by the season of freshening.

II. THE BIRTH WEIGHTS OF CALVES

In spite of the fact that many feeding trials have been conducted with dairy calves, few reports are to be found regarding the birth weights of calves and the factors that influence them. The few reports available however contain data on several hundred animals and so are of considerable value, except in the case of some breeds.

RÉSUMÉ OF PREVIOUS WORK

The work so far reported on this subject can be very easily summarized as Henry and Morrison (3) reported the work available from all experiment stations while later Eckles (1) published data in similar form.

Eckles (1) noted that the first and second calves of heifers were somewhat heavier than those born at later ages but that small calves were often delivered after short gestation periods and large calves after long periods, but between these extremes

TABLE 5
Reported average birth weights of dairy calves

BREED	NUMBER OF CALVES	AVERAGE WEIGHT			RELATION OF WEIGHT OF CALF TO WEIGHT OF DAM
		Males	Females	Total	
Henry and Morrison (2)					
		<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>per cent</i>
Ayrshire.....	34	77	74	76	7.8
Guernsey.....	57	75	68	71	7.1
Holstein.....	104	94	85	89	7.7
Jersey.....	119	58	49	55	6.1
Eckles (1)					
Ayrshire.....	53	73	65	69	6.9
Holstein.....	154	93	88	90	8.0
Jersey.....	196	58	53	55	6.5

there was no evidence of correlation between the weight of the calf and the length of the gestation period.

EXPERIMENTAL WORK

A digest has been made of the herd records kept at the Dairy Farm of Iowa State College from December, 1907, to November, 1921, inclusive and the work reported here includes data on 369 calves and is closely connected with the material presented in paper I of this series. Only records on single calves are included, as it was thought best to treat them alone and the number of twins was too small to really justify treatment here.

An attempt has been made to study some of the factors which might be presumed to have some influence on the birth weights of calves.

In the general summary the breeds have been kept separate, and the scrubs separated from the grades and purebreds. A sex distinction has also been made. The scrubs and grades were animals used in investigational work. A considerable

TABLE 6
Average birth weights of calves

BREED	NUM- BER OF COWS	NUMBER OF CALVES			AVERAGE BIRTH WEIGHT		
		Male	Female	Total	Male	Female	Total
Purebreds							
					pounds	pounds	pounds
Ayrshire.....	14	15	20	35	68	65	66
Guernsey.....	25	42	35	77	66	61	64
Holstein.....	28	41	33	74	97	89	94
Jersey.....	31	38	35	73	55	52	54
Grades							
Ayrshire.....	2	2		2	60		60
Guernsey.....	21	22	26	48	68	63	65
Holstein.....	14	20	17	37	83	77	81
Jersey.....	10	7	12	19	55	53	54
Scrubs.....	4	2	2	4	67	51	59
Grades.....	47	51	55	106	72	65	68
Purebreds.....	98	136	123	259	73	66	70
All calves.....	149	189	180	369	72	65	69

number of grades, of the various breeds except the Ayrshire, are included. These grades vary considerably, as some of them are the result of but one cross of purebred bulls and scrub cows, while others are the result of two and three and in one case four crosses of purebred bulls.

On studying the influence of the age of the cow at the time of freshening on the birth weight of the calf, a few records had to be deleted as the age of the cow and the birth weight of the calf were not always available. Consequently, only 366 records

are available for this part of the work. The age divisions are each of one year, for example, the two-year old group includes all cows two years old but under three. Cows from one to sixteen years of age are included.

TABLE 7
Influence of age of cow at freshening on birth weight of calf

AGE OF COW	NUMBER OF CALVES			AVERAGE BIRTH WEIGHT OF CALVES		
	Male	Female	Total	Male	Female	Total
<i>years</i>				<i>pounds</i>	<i>pounds</i>	<i>pounds</i>
1	1	2	3	97	64	75
2	46	39	85	65	64	64
3	41	33	74	75	67	71
4	26	31	57	78	68	73
5	20	20	40	77	69	73
6	11	14	25	70	65	67
7	13	12	25	79	62	71
8	12	10	22	72	73	72
9	4	6	10	67	76	72
10	3	4	7	79	57	66
11	4	3	7	70	55	64
12	4		4	62		62
13	1	2	3	65	54	58
14	1	2	3	60	59	59
15						
16	1		1	60		60

TABLE 8
Influence of weight of cow at freshening on birth weight of calf

WEIGHT OF COW	NUMBER OF CALVES			AVERAGE BIRTH WEIGHT OF CALVES			RELATION OF WEIGHT OF CALF TO WEIGHT OF DAM
	Male	Female	Total	Male	Female	Total	
<i>pounds</i>				<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>per cent</i>
700	5	5	10	57	52	55	7.3
800	7	11	18	58	58	58	6.8
900	25	26	51	64	59	61	6.4
1000	32	26	58	72	60	66	6.3
1100	20	25	45	73	71	72	6.3
1200	14	19	33	82	72	76	6.1
1300	7	10	17	83	86	84	6.2
1400	10	8	18	103	92	98	6.8
1500	5	3	8	101	97	100	6.5

The number of cases in which the relationship of the weight of the cow at freshening to the weight of the calf at birth are more limited than in some of the other studies as in the earlier part of the period from which the records are taken the cows

TABLE 9

Influence of season of freshening on birth weight of calves

MONTH OF FRESHENING	NUMBER OF CALVES			AVERAGE BIRTH WEIGHT OF CALVES		
	Male	Female	Total	Male	Female	Total
				pounds	pounds	pounds
January.....	12	18	30	75	66	70
February.....	20	23	43	74	76	75
March.....	28	23	51	75	67	72
April.....	15	11	26	66	63	64
May.....	12	13	25	71	60	65
June.....	13	10	23	72	58	66
July.....	14	11	25	59	66	62
August.....	11	16	27	72	68	69
September.....	14	17	31	73	61	67
October.....	19	20	39	76	62	69
November.....	17	10	27	74	73	74
December.....	14	8	22	77	69	74

TABLE 10

Influence of length of gestation on birth weight of calves

LENGTH OF GESTATION	NUMBER OF CALVES			AVERAGE BIRTH WEIGHT OF CALVES		
	Male	Female	Total	Male	Female	Total
days				pounds	pounds	pounds
251	15	2	17	75	43	71
261	23	9	32	70	68	70
271	69	80	149	73	65	69
281	69	75	144	73	65	69
291	4	9	13	69	77	74

were not weighed regularly. Through the greater portion of the time, however, the cows were weighed weekly and the last weight before freshening was considered to be the weight of the cow at freshening. This renders 258 records available. The cows were then classified according to weight. Those weighing

700 pounds to 799 pounds were put in the 700-pound group and so on. The weights arrived at in this way were those used in determining the percentage of the live weight of the dam that the weight of the calf represented.

All the records were available for determining the influence of the season of freshening on the birth weight of the calf. These records were arranged by months.

In considering the influence of the length of the gestation period on the birth weight of the calf, the gestation periods have been taken by ten-day groups, for example, gestations of 251 to 260 days have been grouped together. Only 355 records were available for this purpose.

DISCUSSION OF RESULTS

When the purebred calves are separated out according to breeds, it is found that the Holstein calves lead in weight and are followed by the Ayreshire, Guernsey and Jersey calves. The bull calves are heavier than the heifers in each breed. The grades, with the exception of the Ayrshires which are too small in number to be of significance, approximate the purebreds in birth weight. On the average the scrub calves have the lowest birth weight and the purebreds, the highest. When all classes are grouped together, it is found that the average birth weight is 72 pounds for the bull calves, 65 pounds for the heifer calves and 69 pounds for all.

The age of the cow at the time of freshening may have some influence on the birth weight of the calf. The weights for the calves of yearlings are high, due to the fact that one bull coming in this group is well above the average in birth weight. Leaving this class out of consideration however, it would appear that there is some slight rise in the birth weights of the calves as the cows go from two to five years of age; from then on there is a tendency for the calves to be of lower birth weight.

As the cows increase in weight from an average of 750 pounds in the first group to an average of 1550 pounds in the last group the average birth weights of the calves from these cows increase.

This is true for both the males and females and the average of all. In the case of the average the increase was from 55 pounds to 100 pounds. The relation of the birth weight of the calf to the weight of the dam, when expressed as a percentage of the live weight of the dam does not follow this, however. It decreases from 7.3 per cent in the 700-pound cow group to 6.1 per cent in the 1200-pound group and then shows irregular increases among the groups of greater weight.

It cannot be said that there is any very definite variation in the weight of the calves dropped at different seasons. However it does appear that the average birth weights of the calves dropped in the months of April to October, inclusive, are lower than the weights of those dropped at other times. Within those two main periods, however, there are irregular variations from month to month.

The length of the gestation period has little influence on the birth weights of the calves. However, when the gestation is about the average in length, the calves are near the average in birth weight and as the gestation period varies from normal the birth weight of the calves appears to increase slightly.

SUMMARY

After considering the data presented here the following statements may perhaps be made.

1. The average birth weights found for the calves studied were 72 pounds for males, 65 pounds for females, and 69 pounds for all calves.

2. Of the purebred calves the Holsteins were the heaviest and they were followed by the Ayreshires, Guernseys and Jerseys.

3. The scrub calves had the lowest average birth weight and the purebreds, the highest, though the grades approximated the purebreds in weight.

4. As the cows increase in age up to five years, the average birth weight of the calves apparently increases. From then on there is an irregular decrease in the weights of the calves.

5. With an increase in the weight of the cows there is an increase in the birth weight of the calves, though this is not in

direct proportion to the increase in the weight of the cows. It is not possible to determine from the data available just how closely the increase in weight of the cows follows advancing age. The two factors are correlated to a certain extent and it is difficult to determine which one is of more importance.

6. It may be true that calves dropped from April to October, inclusive, are lighter than those dropped during the rest of the year. There are fairly wide variations in the birth weights of the calves dropped in each of those periods however.

7. The length of the gestation period has little influence on the birth weight of calves, but the more closely the gestation approaches normal in length the nearer are the birth weights to normal. As the gestations recede from normal the greater become the birth weights of the calves.

III. THE RATE OF GROWTH OF DAIRY HEIFERS

In investigational work it is frequently found that a new unit of measurement, with which the results obtained can be compared, is necessary. In view of this fact it was deemed essential that curves for various forms of growth in calves be obtained. Such curves obtained from measurements of animals fed and grown out under normal conditions are extremely useful as bases with which to compare the growth of animals under experimental conditions. Little work of this character is at present available.

RÉSUMÉ OF PREVIOUS WORK

Practically the only work reported on the rates of growth of heifers is that by Eckles (7) who worked with Ayrshires, Holsteins and Jerseys and there the rate of growth to two years of age, when expressed as a percentage of the birth weights and measurements appears to be most rapid in the Jerseys with the Ayrshires second. This confirms general observations regarding the more rapid development of the smaller breeds of dairy cattle.

No effort will be made here to discuss the many factors which control growth but it may be noted that Waters (10) on feeding

calves from the time of birth on rations of various types, from scanty to full-fed, found that in the early stages of development growth in height was more rapid than growth in width. Later however, the growth in width tended to become more rapid. Eckles (6) confirms this observation that the growth impulse is decidedly stronger in the skeleton than in the fleshy parts of the body.

The results obtained by McCandlish (8) on feeding an unsuitable ration to young calves tend to give evidence in the same direction when the increases in live weight and body measurements are considered. The normal animals with which the abnormally fed individuals were compared showed the greatest percentage increase in live weight and then in the body measurements taken. The experimental animals showed greater percentage increases in live weight than in body measurements, but it was found that the decrease in percentage gain in live weight from the normal increase was greater than the average decrease in percentage gains in body measurements. To state it specifically, the percentage gain in live weight was only 30 per cent of normal, while the majority of the body dimensions increased at 50 per cent to 60 per cent of the normal rate. This would indicate that while under normal conditions gains in body weight are more rapid than body measurements, yet under adverse conditions the stimuli which cause increase in skeletal dimensions are not retarded so easily as are the stimuli which induce increases in live weight.

A factor of note in post-natal growth is the decrease in weight which is sometimes said to occur normally for some time after birth. Abnormal decreases are of course to be found in the case of many animals which are not properly nourished or are stunted in some way at that time. No reports of this character have been noted with calves. Robertson (9) however, has reported on the average loss of weight of South Australian infants during the first week after birth. He states that the post-natal loss of weight is due to the mechanical shock at birth, and to the nutritional changes taking place at that time. He finds that the average loss due to these factors is 9.2 per cent of the birth weight.

Post-natal loss is sometimes attributed to the fact that the young do not receive a sufficient supply of nutrients during the first few days after birth. This can hardly be looked on as the sole cause of the loss however, as there is post-natal loss in weight in guinea pigs, which have reached such a stage of development at birth that they can readily forage for themselves soon after that time.

EXPERIMENTAL WORK

The data reported here has been collected from a group of 40 heifers that have been raised from birth to producing age on the

TABLE 11
Change in weight of calves for ten days after birth

CALF NUMBER	SEX	AGE, DAYS										
		Birth	1	2	3	4	5	6	7	8	9	10
		lbs.	lbs.	lbs.	lbs.	lbs.	lbs.	lbs.	lbs.	lbs.	lbs.	lbs.
381	M	83	87	91	94	91	85	87	89	88	88	91
382	M	52	54	55	55	58	55	54	55	57	55	53
384	F	64	62	65	63	61	59	59	59	60	60	61
386	M	108	106	104	100	97	97	102	102	101	99	99
387	M	70	73	71	67	65	65	60	59	60	62	63
Average.....		77	80	80	79	78	76	76	76	77	76	77
Percentage of birth weight.....		100	104	104	103	103	99	99	99	100	99	100

Iowa State College Dairy Farm. These heifers are part of those discussed in section II of this series. The chief reason for obtaining the data was to have a growth curve to use as a basis with which the rates of growth of animals used in experimental work might be compared. The data was collected in the years 1916 to 1920 inclusive.

Purebreds and grades of the Guernsey, Jersey and Holstein breeds, and purebred Ayrshires were used in this work but so far no attempt has been made to arrive at breed characteristics, as the numbers of animals in the various groups at present would be too small for such purposes. The animals have been grouped however, according to the season of the year at which they were

dropped. Winter calves include those dropped from October 1 to March 31 and summer calves are those dropped from April 1 to September 30 inclusive. There are 24 heifers in the winter and 16 in the summer group. All of the heifers were reared under general herd conditions.

TABLE 12

Average live weights and body measurements of winter calves

AGE	LIVE WEIGHT	HEIGHT	DEPTH	WIDTH	PERCENTAGE INCREASE			
					Live weight	Height	Depth	Width
<i>months</i>	<i>pounds</i>	<i>inches</i>	<i>inches</i>	<i>inches</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
Birth	68							
1	91	28.7	11.6	6.9	34			
2	120	30.7	12.6	7.5	76	7	8	9
3	163	32.9	14.2	8.5	140	11	22	23
4	210	34.8	15.4	9.4	209	21	33	36
5	262	36.4	16.7	10.2	285	27	44	48
6	317	38.4	17.9	11.0	366	34	54	59
7	373	39.8	18.7	11.8	449	39	61	71
8	424	41.0	19.7	12.8	524	43	70	86
9	467	42.2	20.3	13.4	587	47	75	94
10	500	42.9	20.9	13.8	635	49	80	100
11	533	43.7	21.5	14.2	684	51	85	106
12	569	44.5	22.0	14.8	737	55	90	114
13	604	45.1	22.6	15.3	788	57	95	122
14	644	45.7	22.8	15.6	847	59	97	126
15	680	46.1	23.4	15.9	900	60	102	130
16	708	46.5	23.8	16.3	941	62	105	136
17	729	46.9	24.0	16.5	972	63	107	139
18	750	47.3	24.2	16.7	1003	65	109	142
19	777	47.7	24.4	17.1	1043	66	110	148
20	806	47.9	24.8	17.5	1085	67	114	154
21	834	48.3	25.2	17.7	1126	68	117	157
22	861	48.5	25.4	17.9	1166	69	119	159
23	894	48.9	25.6	18.1	1215	70	121	162
24	922	48.9	25.8	18.5	1256	70	122	168
29	1010	49.4	26.4	19.5	1385	72	128	183

In addition to the general piece of work, consideration may be given here to results that were obtained on weighing a few calves daily for ten days after birth. In this limited test one heifer and four bull calves were used and the results obtained with them are tabulated.

The general scheme for obtaining body measurements and weights of the heifers used in the major portion of this work can be outlined shortly. Each heifer was weighed at birth. Then on the three consecutive days at the middle of each calen-

TABLE 13

Average live weights and body measurements of summer calves

AGE	LIVE WEIGHT	HEIGHT	DEPTH	WIDTH	PERCENTAGE INCREASE			
					Live weight	Height	Depth	Width
<i>months</i>	<i>pounds</i>	<i>inches</i>	<i>inches</i>	<i>inches</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
Birth	64							
1	89	28.7	11.4	6.3	39			
2	119	30.7	12.8	7.3	86	7	12	16
3	163	32.7	14.4	8.3	155	14	26	32
4	212	34.6	15.7	9.4	231	21	38	49
5	262	36.4	16.9	10.2	309	27	48	62
6	309	38.2	17.9	11.4	383	33	57	81
7	355	39.8	18.7	11.8	455	39	64	87
8	396	40.8	19.5	12.4	519	42	71	97
9	429	41.8	20.1	13.0	570	46	76	106
10	464	42.5	20.7	13.4	625	48	82	113
11	504	43.3	21.3	14.0	688	51	87	122
12	540	43.9	21.7	14.4	744	53	90	129
13	576	44.5	22.2	14.8	800	55	95	135
14	607	44.9	22.6	15.3	848	56	98	143
15	629	45.3	23.0	15.4	883	58	102	144
16	651	45.9	23.2	15.7	917	60	104	149
17	676	46.5	23.6	15.9	956	62	107	152
18	698	46.9	24.0	16.3	991	63	111	159
19	720	47.3	24.2	16.5	1025	65	112	162
20	749	47.5	24.6	16.7	1070	66	116	165
21	782	47.7	24.6	17.1	1122	66	116	171
22	815	47.9	25.0	17.5	1173	67	119	178
23	844	48.3	25.4	17.5	1219	68	123	181
24	874	48.5	25.4	17.9	1266	69	123	184
29	941	49.2	26.2	18.9	1370	71	129	200

dar month all the heifers were weighed and on one of these days the body measurements were taken. This method was pursued to avoid continuous weighing of the animals. The three consecutive weighings obtained were averaged and then the average of the first series and the average of the second series of weigh-

ings were averaged to obtain the weights of the animals at one month of age. This method was then followed through to the time of freshening. It is believed that it gives a reasonably true average as at the first weighing the animals would range up to

TABLE 14
Average live weights and body measurements of all calves

AGE	LIVE WEIGHT	HEIGHT	DEPTH	WIDTH	PERCENTAGE INCREASE			
					Live weight	Height	Depth	width
<i>months</i>	<i>pounds</i>	<i>inches</i>	<i>inches</i>	<i>inches</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
Birth	67							
1	90	28.7	11.4	6.7	34			
2	120	30.7	12.8	7.5	79	7	11	12
3	163	32.9	14.2	8.5	143	15	25	27
4	211	34.8	15.4	9.4	216	21	35	40
5	262	36.4	16.7	10.2	291	27	47	52
6	314	38.4	17.9	11.2	369	34	57	67
7	366	39.8	18.7	11.8	446	39	64	76
8	413	41.0	19.5	12.6	516	43	71	88
9	452	42.0	20.3	13.2	575	46	78	97
10	486	42.7	20.9	13.6	625	49	83	103
11	521	43.5	21.5	14.2	677	52	89	112
12	557	44.3	21.9	14.6	731	54	92	118
13	592	44.9	22.4	15.1	784	56	96	125
14	628	45.5	22.8	15.4	837	59	100	130
15	659	45.9	23.2	15.7	884	60	104	134
16	685	46.3	23.6	16.1	922	61	107	140
17	708	46.7	23.8	16.3	957	63	109	143
18	729	47.1	24.2	16.5	988	64	112	146
19	754	47.5	24.4	16.9	1025	65	114	152
20	784	47.7	24.6	17.1	1070	66	116	155
21	813	48.1	25.0	17.3	1113	68	119	158
22	852	48.3	25.2	17.7	1171	68	121	164
23	874	48.5	25.4	18.1	1204	69	123	170
24	903	48.9	25.6	18.7	1248	70	125	179
29	982	49.4	26.4	19.3	1366	72	132	188

30 days of age and at the second from thirty to sixty days of age and the average of all would be thirty days of age.

The body measurements taken were the height at withers, depth of chest and width at hooks. These measurements were averaged in the same way as were the live weights. No measurements were taken at birth however.

DISCUSSION OF RESULTS

It will be noted in the tabulation that the live weights, starting at one month of age, are tabulated by months until the age of two years is reached. After that age the heifers were dropped from the work as they freshened and the live weights and body measurements are consequently omitted after that with the exception of the averages for the animals at the time of freshening. The average age of freshening was 29 months for each group.

TABLE 15

Ratio of average live weight of calves in pounds to product of average height, depth and width in inches

AGE	RATIO 1:	AGE	RATIO 1:
<i>months</i>		<i>months</i>	
1	24.5	14	25.4
2	24.6	15	25.4
3	24.4	16	25.7
4	23.9	17	25.6
5	23.7	18	25.9
6	24.5	19	26.0
7	23.9	20	25.6
8	24.4	21	25.6
9	24.9	22	25.3
10	24.9	23	25.5
11	25.9	24	25.9
12	25.4	29	25.6
13	25.7		

In surveying the results obtained on weighing calves daily for 10 days after birth it may be said that immediately after birth the calves appear to gain 4 per cent in live weight, probably due to intestinal fill and then gradually decrease until they have reached only 1 per cent below their birth weights and then come back to normal. These results do not indicate any marked changes in live weight immediately following birth, though there are greater changes in individual cases.

The winter calves averaged 68 pounds in live weight at birth, the summer calves, 64 pounds, and the total 67 pounds. This does not introduce any variation of importance.

There are variations from time to time in the rates of development of the two groups of heifers but at the age of two years the winter heifers exceed the summer heifers in all body measurements and in live weights, but have excelled the summer heifers in only one increase—the percentage increase in height. By the time the age of freshening is reached however, the winter heifers are excelled only in the percentage increase in depth of chest and width of hocks. The significance of this is probably not great, especially as the winter heifers were larger and heavier than the summer heifers at the time of freshening.

Formulae have at various times been presented for the calculation of the live weight of various classes of live stock from body dimensions. Without discussing the accuracy or practicability of such methods a table is presented here which shows that from the data obtained in this work, it was observed that there was a fairly constant relationship between the live weight in pounds and the product of the height, depth and width in inches. This ratio is approximately 1:25. It holds fairly well for the averages of the group but would vary considerably with individuals. There also appears to be a variation connected with the ages of the animals as with the younger animals the ratio tended to become narrower while it widened with the older individuals.

SUMMARY

From the data presented here a few facts appear to be in evidence.

1. There is apparently little indication of a post-natal loss in the weight of calves.
2. The live weight of the animals shows the most rapid increase.
3. When the body measurements taken are considered it is found that in rapidity of increase their rank from the highest to the lowest is width, depth and height.
4. Between winter and summer heifers there appears to be little difference except that the winter heifers reach a greater weight and greater body measurements by the time of freshening than do the summer heifers.

5. There appears to be a fairly definite ratio between the live weight of the animals in pounds and the product of the height, depth and width in inches.

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REVIEW OF FOREIGN DAIRY LITERATURE

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INTRODUCTION

In my survey of French dairy literature, I shall attempt to bring before the readers of the JOURNAL, the current thought of French dairy interests both from the scientific and practical standpoints by means of abstracts of articles as they appear in French publications. At times I shall offer complete translations if they seem to be of particular interest, to a large group or, if an especially good piece of work is published.

The following papers among others will be reviewed:

1. *Le Lait*, a general review of matters pertaining to the dairy industry, published monthly at Lyon.
2. *La Laiterie*, a paper published semi-monthly in Paris devoted to the interests of the dairy farmer and creameryman.
3. *Lacta*, a world wide review of the dairy industry published in Paris.
4. *L'industrie Laitiere*, official bulletin of the French Society for the encouragement of the dairy industry.

I shall be glad at any time to cover subject matter that may be suggested by the reader.

DRUGE, F., Chemist of the Maison, Lefevre, Utile at Nantes. Two short investigations on milk. *Le Lait*, 2nd year, February, 1922, no. 2.

Influence of Chloroform and of toluene on the activity of rennet

The milk used in the experiment was fresh mornings milk. It was immediately distributed in three Erlenmeyer flasks marked with the letters A, C, and T.

Bottle A contained pure milk.

Bottle C chloroform added at the rate of 14 grams per liter.

Bottle T toluene added in the same proportions, 14 grams per liter.

The initial acidity of the milk was equal to 155 cc. of soda $\frac{N}{10}$ per liter equivalent to 1.39 of lactic acid.

Each of the flasks was then rapidly raised to a temperature of 35° and one milligram of rennet for each 100 cc. of milk added. The temperature was maintained at 34–35° until the end of the experiment.

The rapidity of coagulation shows the following:

Bottle A, coagulation complete and firm in 87 minutes.¹

Bottle C, coagulation complete and firm in 122 minutes.¹

Bottle T, coagulation complete and firm in 92 minutes.¹

Chloroform retards the activity of rennet to a considerable extent; toluene has an insignificant effect.

Influence of chloroform and of toluene on the spontaneous coagulation of cow's milk

This experiment has been carried on under the same conditions as in experiment I. The bottles have however been doubled, i.e., we had two series of flasks A, C, and T.

The first series was left at laboratory temperature about 18°C. The second was put in a cold cellar at 50°C.

The initial acidity, titrated according to the usual procedure, was practically 1.39 lactic acid per liter. The following results were obtained.

	AFTER 24 HOURS	AFTER 24 HOURS	AFTER 48 HOURS	AFTER 72 HOURS	AFTER 96 HOURS	AFTER 120 HOURS
	grams	grams	grams	grams	grams	grams
Bottle A {	+18°C....	1.48	1.495	5.85	7.85	5.13
	+5°C....			Coagu- lated	Coagu- lated	In filtered serum
		1.39	1.39	1.39	1.39	1.59
Bottle C {	+18°C....	1.44	1.44	1.44	2.11	4.96
	+5°C....					In filtered serum
		1.39	1.39	1.39	1.39	1.39
Bottle T {	+18°C....	1.44	1.44	1.44	4.95	4.95
	+5°C....				Coagu- lated	In filtered serum
		1.39	1.39	1.39	1.39	1.87

¹ Lactic acid per liter of serum resulting from the filtration of the coagulated milk: Ogr. 90.

The inhibiting action of chloroform on the lactic ferments is now unquestionable; that of toluene is much more feeble. In combining the action of low temperature and that of chloroform, milk may be preserved for five days without its acidity increasing, a great advantage in certain kinds of investigation.

BLIN, HENRI, Laureat de l'Academie d'Agriculture. Cooperative creameries in Brittany. *La Laiterie*, January 21, 1922, vol. 28, no. 2.

Brittany with its temperate climate, pastures, and its facilities for export of butter to England, a great consuming country, offers outstanding advantages for dairy development. M. Blin says there is a great opportunity for the development of the butter industry in this northwestern section of France. He points out that Denmark, hardly as large as Brittany and Normandy combined, with its small farms by means of cooperative organization, has been able to hold first place on the English market. Specialization has made the Danes watch closely the functioning of their coöperatives to keep down the cost of production. The sale of their products is a vital necessity, so that they manufacture on a large scale a product of a superior quality.

Denmark has more than 1500 dairy coöperatives with more than 150,000 members exporting to England 90,000 metric tons, while France ships hardly 20,000 (pre-war figure). Ireland has federations organized on coöperative principles while Germany has several thousand. In France there were before the war, 106 coöperative dairies in Charentes and Poitou with 60,000 members making 8,000,000 kgm. of butter annually, valued at nearly 12,000,000 francs.

The writer points out that there is no reason why Brittany should not follow this progressive movement and that it will be a great day for the industry when the Briton farmers have been convinced of the advantages of cooperation. Private capital shows itself capable of operating creameries on a large scale. M. Blin believes it just as easy for farmers to operate creameries coöperatively if they will only analyze the situation more closely and work together with the proper spirit. He shows that private enterprise at the present time is taking advantage of the unorganized French farmer and that it is time for concerted action.

He reviews the production of the different provinces as reported in 1914. In la Loire-Inferieure there were 182,956 dairy cows producing 1,452,132 hectoliters of milk. The production of butter approximated

400,000 kilos annually. In the department or province of Ille et Vilaine, which ranks first as a province in the production of milk, there were 433,000 cows producing 4,521,347 hectoliters of milk. Finistère with 260,000 cows produced 1,408,500 hectoliters of milk and 312,130 kgm. of butter were made. Morbihan with 212,421 cows produced 2,727,984 hectoliters of milk. Côtes-du Nord which ranks fourth in production of milk in France has a cow population about equal to Morbihan and produces 3,000,000 hectoliters of milk. In Ille and Vilaine there are some commercial creameries and a few in Côtes-du-Nord. However the coöperative creamery is practically unknown except for a little development in Morbihan.

M. Blin emphasizes the advantages of the coöperative creamery. They are able to reduce the cost of making and to make a standardized product of superior quality. Further, the use of centrifugal separators means greater efficiency in the removal of fat from the milk. The French farmers suffer great losses every year by the use of the shallow pan method of skimming.

The organization of the coöperative creamery calls for a capitalization of from 40-50,000 francs of which 20-22,000 francs is required for equipment. There must be 200 to 250 members or patrons furnishing 200 francs besides being able to supply a total of 4-6000 liters of milk daily. M. Blin shows how the financing may be taken care of by brokerage on the milk consigned by each patron. In conclusion he says: "There is a place in Brittany for coöperative creameries, assuring a large remuneration for the efforts expended and the capital invested. Rural economy, in this country, ought to find a productive source of the most happy results for the small producers in the exploitation of the industrial creamery under the guarantee of the coöperative organization."

S. H. H.

LUCAS, J. E. AND LEROY ANDRÉ. The Food Value of Milk with Reference to its Sale Price. *Le Lait*, 2nd year, January, 1922, no. 1.

The problem of supplying milk to the centres of large population in France has for several years with the beginning of each winter, become more momentous. Lucas and Leroy point out that such a situation is prejudicial to the public health since milk is an ideal food for children, the sick, the aged and as well for adults; that it deserves a more important place than that which it occupies because of its high nutritive

value and its desirable physiological effects on the human body. Civil authorities and children's aid societies have interested themselves in this serious question to find out the causes and remedies.

In spite of the rise in price of milk during the war to 1 franc 10 centimes (normal exchange 22 cents) it is with the greatest difficulty that people in Paris and its suburbs supply themselves with milk especially in the winter season. The normal price of milk for the winter of 1913 was 40 centimes (8 cents). In 1914 the daily rail shipments of milk into Paris amounted to 900,000 litres while those of 1919 reached barely 560,000 litres and that to supply a population increased by 350,000.

The writers say this shortage of milk is due to such things as the cost of raising dairy cows, the difficulty of procuring capable help, market price of hay with plenty of demand for it, the ready market for butter and cheese, and lastly, unjustifiable legal suits against dairymen.

It requires about 9 francs per day to feed a cow in the vicinity of Paris, this figure including feed costs, labor, interest on investment and the items of so-called indirect or overhead expense. This makes a farm cost of 90 centimes per litre. Add to this transportation, distribution, and plant charges and there is no margin of profit left even with milk selling at 1 franc 10 centimes. This they say would discourage production.

Further the authors point out that it is hard to get help on dairy farms because of the long hours. "The painful conditions of his (the herdsman) existence are not in harmony with the desires for gain and of easy life which are getting into the rural masses."

The price offered for straw and hay on the large city and country markets is high enough to discourage its being fed to milch cows. Further milk has been severely taxed while butter and cheese have not. The farmer at a distance from a large city has found it more advantageous to manufacture his milk into cheese or butter, keeping the skimmilk and buttermilk on the farm for feeding pigs.

Legislative acts have prevented in many places the rise in price of milk to meet the cost of production. As a result, a great many dairymen have been driven out of business. In conclusion, Lucos and Leroy believe that the principal reason for the milk crisis has been the suppression by acts of the government of the normal functioning of the law of supply and demand so that the price of milk has not kept pace with the cost of production and its food value.

In a table presented, the food value of milk is compared with that of meats, vegetables, eggs, chocolate, etc. The superior value of the amino acids in milk is discussed as compared with those of other foods. "The vitamins, or accessory factors of growth have an unknown chemical nature which exists in the majority of our foods and whose presence in our rations is indispensable to the best condition of the body." Milk is emphasized as a protective food from the health standpoint containing food elements necessary to maximum growth and the most healthy condition of the body. Further they point out that from the energy standpoint alone that "in the many modest budgets notable economies could be realized by replacing a certain portion of the meat in the ration by an isodynamic quantity of milk or milk products."

The problem in France, particularly in the large cities, is to obtain a larger milk supply. Lucas and Leroy conclude "that it is only by an increase in milk production on the farms that a satisfactory supply in the large cities will be insured. Good measures will be those which stimulate the stockmen permitting them to realize the maximum of profit. It is by the return to normal functioning of economic laws, by the total suppression of taxation, that a return to normal will be obtained, that is to say, a sufficient market supply. Complete liberty must be returned to the milk business and a higher price paid for its product for some time."

Comment. France has been suffering from a shortage of milk during and since the war period and the supply is not getting back to normal. Health authorities see that it is a health problem and are trying to foster measures that will stimulate its production. Dairy farmers in France seem to have their troubles as well as in America, though perhaps from a slightly different viewpoint.

S. H. H.

BOOK REVIEW

The Marketing of Whole Milk by Henry E. Erdman is a recent addition to the Citizens Library of Economics, Politics and Sociology, edited by Dr. Richard Ely and published by the MacMillan Company.

After an introductory chapter, the presentation is organized around the headings of milk as a community commodity, the market for whole milk, distribution of milk, collective bargaining in the sale of whole milk, milk prices, consideration of proposed remedies, and conclusions.

Dr. Erdman approaches the milk problem primarily from the standpoint of the economist but his presentation makes evident an intimate and sympathetic acquaintance with the complex details of the business. He appreciates the desirability of grades of milk and the necessity of standardization in connection with buying milk on the basis of its food value. The place of the middleman is pointed out, the limitations of centralization of milk companies are noted and the question of duplication in milk delivery is well presented.

The handling of the various economic aspects of the milk question is particularly good.

Taken all in all this is perhaps the simplest and clearest available presentation of the manifold problems of the city milk business. The form of presentation, while concise, will appeal to the layman as well as to the student of the subject. There is a good table of contents and subject index and the mechanical work is up to the high standard of the publishers.

H. A. H.

ANNOUNCEMENTS

FOREIGN DAIRY LITERATURE. Beginning with this issue of the *JOURNAL OF DAIRY SCIENCE*, the first effort is made to give reviews and abstracts of dairy literature appearing in foreign publications. Prof. S. H. Harvey of College Park, Maryland, has generously volunteered to review and abstract dairy journals, bulletins and other French publications of interest to our readers. Plans are being developed for the regular abstracting of dairy literature from other foreign countries. This, of course, is a big task and can only be accomplished by having the hearty coöperation of all members of the American Dairy Science Association qualified for this work. We hope that you will volunteer service in this connection and that you will give us your suggestions regarding other persons particularly qualified for this kind of work.

NEWS NOTES. Many of the readers of the *JOURNAL* are particularly interested in news items such as have been running from time to time under the heading "Dairy Notes." This department can be kept up only if each member will coöperate by sending in such news items as come to his attention. Please feel free to send such items to any member of the editorial board, but preferably to Secretary J. B. Fitch.

CONDENSE YOUR PAPERS. During the last few months there has been a remarkable increase in the number of papers submitted for publication in the *JOURNAL OF DAIRY SCIENCE*. This of course means that on account of the limited size of the *JOURNAL*, some authors have been disappointed. The problem of how to provide space for all worthy papers can be solved at least in part, if each author will coöperate by condensing his paper as much as he possibly can without sacrificing any of the essential features of his message. Your hearty coöperation in this matter is earnestly solicited.—J. H. FRANDSEN, *Editor*.

DAIRY NOTES

The Dairy Division of the United States Department of Agriculture reports the following resignation:

E. V. Ellington, after approximately eight years of service in the Dairy Division, B. A. I., resigned on March 6 to accept the position of professor of dairying at Washington State College and head of the dairy division of the experiment station at Pullman, Wash. His successor has not been selected. He was formerly acting in charge of the Dairy Extension Section, in which capacity he directed extension work with cow-testing and bull associations, cheese manufacturing, and general dairy farming, and was recently made associate in charge of dairy introduction work.

The following changes are reported by the Dairy Department of the Connecticut Agricultural College at Storrs, Connecticut:

Prof. P. A. Campbell, who has been extension dairyman for two years, has left the department to take charge of a large dairy farm near Philadelphia, Pa. *Mr. A. H. Merrill*, who succeeds Mr. Campbell, has for the last two years been manager of a large stock farm at Dixville Notch, N. H. He previously had taught dairying in two of the leading secondary schools of agriculture in this part of the country. He is a graduate of the New Hampshire State College.

Mr. J. A. Simms, who has been in general live stock work other than dairy cattle with the extension service, has been appointed as an assistant to Professor Merrill. Mr. Simms is a graduate of North Carolina Agricultural College.

Washington State College at Pullman, Wash., reports the resignation of *E. G. Woodward* as head of the dairy department. Mr. Woodward resigned to go into commercial work.

THE CULTURE COLLECTION OF THE SOCIETY OF AMERICAN BACTERIOLOGISTS

The Society of American Bacteriologists has taken over the collection of cultures which for the past ten years has been maintained at the American Museum of Natural History by Prof. C.-E. A. Winslow,

and has deposited it at the Army Medical Museum, where facilities have been arranged for its housing and maintenance.

The following committee will be in charge:

Major G. R. Callender, Curator of the Army Medical Museum
Dr. Geo. W. McCoy, Director Hygienic Laboratory, U. S. Public Health Service
Major H. J. Nichols, Army Medical School
The President of the Society
The Secretary of the Society
Dr. J. M. Sherman, Dairy Division, Bureau of Animal Industry, Chairman

These and other members of the Society in and near Washington will do volunteer work and the Research Fellow will do part time work in maintaining the collection. No charge will be made for cultures. In making requests, the classification of the Society should be followed as far as possible.

Mail should be addressed to the Department of Bacteriology, Army Medical Museum, 7th and B Streets, S. W., Washington, D. C.

J. M. SHERMAN,
Chairman of the Committee.

A RESEARCH FELLOWSHIP IN BACTERIOLOGY

It is announced by Dr. Victor C. Vaughan of the National Research council in Washington, that the Society of American Bacteriologists at its recent meeting in Philadelphia, appropriated a fund for the support of a Research Fellowship in pure bacteriology. While excellent work is being carried on in many places, nearly all the problems under investigation have as their aim a practical application and there are, therefore, many gaps in our knowledge of fundamental principles. The Society, believing it to be the duty of bacteriologists to fill these *lacunae* requires that the line of work to be carried on under its fund must concern a purely scientific and fundamental phase of bacteriology, although a certain latitude of choice will be permitted, conditioned by the previous training and the desires of the Research Fellow himself.

Applicants for the fellowship must have the degree of B.S., or its equivalent. The successful candidate, through arrangements now being made, will receive academic credit for the work done from a university of recognized standing. One hundred dollars a month will be available for the living expenses of the fellow. Approximately half his time will be devoted to details connected with the Society's collection of bacteria, deposited at the Army Medical Museum.

The selection of the Research Fellow will be in charge of a committee consisting of:

Dr. Victor C. Vaughan, Chairman Medical Section, National Research Council; Chairman

Captain C. S. Butler, Medical Corps, U. S. Navy, Commandant, Naval Medical School

Dr. Geo. W. McCoy, Director Hygienic Laboratory, U. S. Public Health Service

Dr. John R. Mohler, Chief, Bureau of Animal Industry

Mr. L. A. Rogers, (President of the Society of American Bacteriologists) In Charge of Research Laboratory, Dairy Division, Bureau of Animal Industry

Colonel Joseph F. Siler, Medical Corps, U. S. Army, Division of Sanitation, Office of the Surgeon General of the Army

Dr. Erwin F. Smith, Pathologist in Charge, Laboratory of Plant Pathology, Bureau of Plant Pathology

This committee will have general supervision of the work, approve the problem selected and pass upon the thesis which the fellow will submit as the report of his research.

Applications for and communications concerning the Research Fellowship should be addressed to the Chairman of the Committee, Dr. Victor C. Vaughan, National Research Council, Washington, D. C.

A. PARKER HITCHENS, Major, M. C.

Secretary of the Committee.

*Army Medical School,
Washington, D. C.*

A COMPARISON OF EARLY, MEDIUM AND LATE MATURING VARIETIES OF SILAGE CORN FOR MILK PRODUCTION¹

A REPORT OF PROGRESS

GEORGE C. WHITE AND LEROY M. CHAPMAN

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Connecticut*

The data herein reported represents the first year of an investigation that is planned to continue for at least three years, and are, therefore, to be regarded as of a preliminary nature. However, some light seems to be thrown upon a debatable problem and it is the hope of the authors that criticisms and comment will be aroused among those interested.

THE PROBLEM STATED

Silage is an important crop throughout the dairy sections of the country. In New England and the northern tier of states, where the season is short, the tendency has been to grow special varieties for silage, the seed for which is secured from more southern states producing a large green weight per acre. This has been particularly true in regions where tillable land is limited and farmers purchase much of the grain fed. On the other hand, many farmers in this region believe that earlier strains of corn are more satisfactory for silage.

It should be clearly held in mind that the problem is not "when to harvest any given variety for silage," which was formerly much discussed in the states of longer growing season. The work of several stations, particularly that of the Indiana Station (1) definitely settled that point. In the case now under discussion, the farmer must choose between a large, late maturing

¹ Contribution from the Storrs Agricultural Experiment Station.

variety that will give high total yield, but with the ears very immature, and an earlier, smaller corn that yields less tonnage but has better matured ears.

In 1914, field trials of varieties and strains of corn were begun by the agronomy department. Early results indicated that late varieties did not yield as much *dry matter* as those that would practically mature, but as data accumulated this has proven to be incorrect as shown by the table below.

Considering merely the yields of green and dry matter per acre, it would seem logical, therefore, to grow a late variety. An attempt was made to compute the feeding value of the several

TABLE 1
Yields per acre of silage corn (average of five years)

STAGE OF MATURITY	GREEN WEIGHT	DRY MATTER	WATER
	<i>pounds</i>	<i>pounds</i>	<i>per cent</i>
Early group.....	24774	6491	73 80
Hard dough.....			
Medium group.....	32947	7218	78 09
Soft dough.....			
Late group.....	40648	8064	80.16
Kernels just forming.....			

It should be borne in mind that these are distinct groups of varieties, all planted and harvested at the same time, and that they represent the range found in actual practice in New England.

silages on the basis of digestion coefficients available through the work of Lindsey and Armsby, and to use results in an equation that would include the yield per acre. This proved fruitless because, first, the coefficients are given simply as for "mature" and "immature" corn and second, these coefficients do not throw sufficient light on the "milk-producing value" of the several feeds.

It was then decided to undertake a feeding investigation to answer the following questions:

1. What is the value per ton of early, medium and late types of silage corn for milk production?

2. What is the acre value of these several types for milk production?

PLAN OF THE EXPERIMENT

The silos

Three small silos, 36 × 6 feet, were purchased and erected during the summer of 1920. They are of spruce staves, set on a concrete base without drains and are covered with separate conical, metal roofs. The inside surface is treated with a wood preserver supplied by the manufacturer of the silos.

The diameter is small to allow feeding to a small group of cows without spoilage. Normal silage can be produced in very small containers as has been shown by Eckles et al. (2), Newton (3), and Westover and Garver (4). No difficulty was met in this respect, excellent silage resulting in all cases.

The silage

Three types of corn were used, early (Pride of the North), medium (Leaming) and late (Eureka). These were planted on May 28, 1920, as part of a large field of corn on the college farm and at cutting time, October 4, were sampled and analyzed with the following results:

TABLE 2

SILO-VARIETY	STAGE OF MATURITY	ANALYSIS IN PERCENTAGE							
		Water	Dry matter	Ash	Crude protein	Crude fiber	N-free extract	Fat	
A. Pride of the North (early).....	Ripe, basal leaves dry	70 22	29 78	1 17	2 48	5 87	19 44	0 81	
B. Leaming (medium) . . .	Soft dough	75 04	24 96	0 99	1 92	5 33	16 16	0 56	
C. Eureka (late) . . .	Early milk	79 49	20 51	0 89	1 28	5 95	11 97	0 29	

It will be noted that the corn used for filling these silos is somewhat higher in dry matter and lower in per cent water than the five year averages for the types given in table 1, due largely to season. During the three weeks following filling of the silos, the one containing the late corn lost considerable juice, the medium lost very little and the early practically none.

The silage feeding

Silage, in the usual ration, is only one of the food components and comprises, in nutrients supplied, approximately one third of the total amount consumed. Other investigators who were consulted, while planning this experiment, felt that the difference

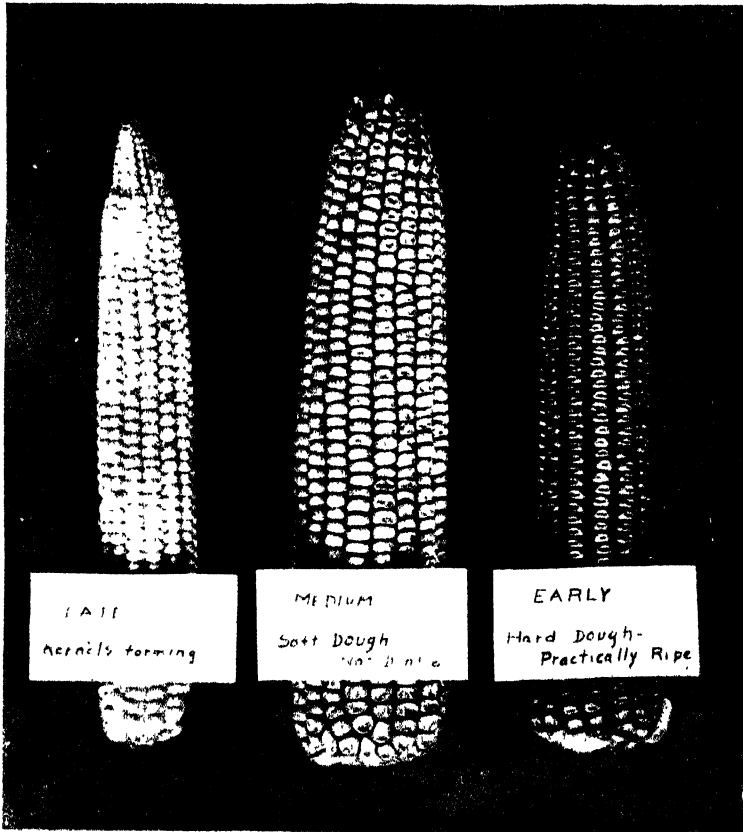


FIG. 1. LEFT TO RIGHT. EUREKA, LEAMING AND PRIDE OF THE NORTH PHOTOGRAPHED AT TIME OF ENSILING

The Leaming in center was not dented although having a suggestion of being dented due to slight loss of moisture before being photographed.

in feeding value of silage of different maturity could not be successfully measured by a feeding trial. To have the silage contribute a considerable proportion of the total nutrients, therefore, seemed a necessity. With this in view the plan adopted called

for the feeding of 50 pounds daily to all animals that were found, in the preliminary trial, capable of eating this amount. One animal in each group was fed only 40 pounds, they being less capable of handling the larger amount cleanly. The feeding of 50 pounds daily, while high, is in line with the practice in a good many herds. In order to eliminate variables, these amounts were fed throughout the experiment.

The silage was removed twice daily from the silos in burlap bags at feeding time, weighed to each animal, and the amount recorded. The amount refused by each cow was weighed and the net weight consumed recorded for each cow. The amount refused was small and nearly identical in all three groups.

The hay feeding

The hay used was all from one uniform lot, a mixture chiefly of timothy with a small amount of redtop and clover. The quality was quite similar to a great deal of New England hay with respect to legume content, and more uniform it is believed, than hay of a higher clover content would have been.

The hay was fed soon after noon each day and four pounds was the amount fed to each animal. The small amount fed enabled the cows to consume a large quantity of silage. There was no hay refused.

The grain feeding

A grain ration that has been widely used for experimental feeding of dairy cows consists of 4 parts cornmeal, 2 parts bran and 1 part oilmeal. This ration was recommended by a committee of the American Dairy Science Association a few years ago. However, it seems better adapted for feeding with alfalfa or clover hay than with mixed hay and it does not contain cottonseed meal which is in common use as a dairy feed in the East.

For this experiment a ration consisting of 3 parts cornmeal, 3 parts wheat bran, and 2 parts cottonseed meal (36 per cent protein) seemed best adapted. Such a ration supplies a good amount of protein and provided bulk and palatability; it is quite similar to the general type of New England ration and is not greatly different from the experimental ration referred to.

Applying the digestive coefficients (5) 1 pound contains 0.148 pound digestible protein and 0.7171 pound total digestible nutrients. It has a nutritive ratio of 1 to 3.805. A ration of 50 pounds of silage, 4 pounds of hay and 8 pounds of grain provides a nutritive ratio of 1 to 6.73. The same roughages with 10 pounds of grain provides a ratio of 1 to 6.38; with 12 pounds of grain, 1 to 6.13; and with 6 pounds of grain, 1 to 7.21.

The grain was fed twice daily immediately after the milking periods and preceded the feeding of the silage at about 6:30 a.m. and 5:30 p.m. The daily live weight was used as a guide for feeding the grain. When an animal showed a tendency to gain the grain was reduced either $\frac{1}{16}$ or $\frac{1}{8}$ of a pound and generally followed in two or three days by a further alteration to make a full half pound reduction. In case of loss of weight the procedure was reversed. The grain fed throughout was from one special lot mixed in a commercial plant under the direction of one of the authors.

Salting and watering

Salt was placed before the animals three times each week. Water was placed before them in the forenoon, afternoon and at night.

The analysis of the feeds and milk

The hay was sampled from each fourth bale, 2 pounds being taken from each, after the bale had been broken.

The grain was sampled at the plant from each fifth bag during the process of filling, by the official sampler of the State Experiment Station, at New Haven.

The silage was sampled from the surface of each silo every ten days. A quart fruit jar was filled during the time the silage was being removed from the silo on three consecutive days. The three days samples of each type of silage were mixed and analyzed.

A sample of the morning and night's milk from each cow was taken at ten-day intervals, composited and tested for fat and total solids by means of the Babcock test and lactometer.

The analysis of the feeds is given in the following table:

TABLE 3
*Analysis of feeds **

FEED	WATER	ASH	PROTEIN	FIBER	N-FREE EXTRACT	FAT
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
Hay.....	4 23	5.50	7.25	30.71	50.25	2.06
Mixed ration.....	11.67	3 44	19 44	7 01	53.26	5.18
Bran.....	10 92	5 06	17 44	10.62	50.15	5.81
Corn meal.....	16.00	1.23	8.44	2.06	68 99	3.28
Cottonseed meal.....	8 42	6 26	36.50	11.78	29.98	7.06
Early silage.....	74.26	1.24	2.46	5.87	15.34	0.80
Medium silage.....	74 83	1.30	2 18	5 83	15 27	0.68
Late silage.....	80.57	1.19	1 68	6.84	9.97	0.47

* All of the feed analyses were made by the Chemistry Department of the Connecticut Agricultural Experiment Station at New Haven, to whom our sincere thanks are due.

The silage analyses revealed that the early and the medium were very similar in composition and presumably of about equal feeding value. The medium was uniformly a little lower in protein and fat. In round numbers the late silage compared to the early was 6 per cent higher in water, slightly lower in ash, nearly 1 per cent lower in protein, 1 per cent higher in fiber, 5.5 per cent lower in nitrogen-free extract, and contained approximately half the amount of fat. Theoretically the early should have a very slightly higher feeding value than the medium and about 20 per cent higher feeding value than the late. This assumption was clearly borne out in the feeding trials.

The cows

Data concerning the experimental cows is given in table 4. The age and initial weight is based upon January 25, 1921, this being the date of the beginning of the experiment proper.

The age is given in years and months. All animals in the experiment are pure-bred except Colony. Especial attention was given to balancing the early and late groups as to size and breed.

Polly in the early group and Colony in the late, being immature could not be prevented from gaining in weight. When the grain

was reduced the milk declined rather than the weight. The fact that Polly had calved in August, 1920, and was again bred in October made her especially unsuited to this experiment, the secreting stimulus being at such low ebb that the impulse of growth could not be checked. The data from these two animals were accordingly rejected. Lute gave birth to twins and was considerably upset for some time thereafter. She was, therefore,

TABLE 4
Data concerning experimental cows

NAME	BREED	AGE	GROUP	CALVED	BRED	INITIAL WEIGHT	FINAL WEIGHT	EXTREME VARIATION IN WEIGHT FROM INITIAL	
								Weight	Date
Pansy.....	Holstein	6-10	Early	12/14/20	4/ 7/21	1095	1125	1125	5/4
Polly.....	Holstein	2-11	Early	7/ 1/20	10/10/20	956	1081	1081	5/4
Fruition.....	Holstein	3-11	Early	12/29/20	4/12/20	1160	1161	1124	3/5
Copper Butterfly 2d.....	Jersey	10-0	Early	11/23/20	1/12/21	887	909	911	4/24
Simple.....	Guernsey	9-4	Medium	12/23/20	3/21/21	1030	1039	1056	3/26
Corona.....	Holstein	10-10	Medium	9/20/21	4/ 8/21	946	948	961	3/15
Storrs Robin 2d...	Jersey	8-9	Medium	11/15/20	2/26/21	887	883	908	3/25
Lute*.....	Guernsey	4-5	Medium	1/ 4/21	5/ 7/21				
Filene.....	Holstein	3-10	Late	11/19/20	1/19/21	1059	1052	1066	4/4
Colony.....	Holstein	2-6	Late	11/26/20	3/14/21	915	967	972	4/24
Fusion.....	Holstein	3-11	Late	12/27-20	3/17-21	1292	1248	1228	3/5
V. J. Storrs.....	Jersey	5-9	Late	12/ 8/20	1/28/21	800	809	836	3/26

* Did not start on experiment.

never placed on experiment. As a result the actual results of the experiment are, therefor, the averages of three cows per group.

Both Fruition in the early group and Fusion in the late group, having recently calved, gave some trouble in the fore part of the experiment, as a result of the attempt to hold them at constant weight. It is an interesting fact that both, at one time, were declining in weight due to too heavy feeding of grain, they not

being able to handle 15 or 16 pounds and clean up 50 pounds of silage. The result was a rejection of part of the silage and a decline in milk. A sharp cut of $3\frac{1}{2}$ pounds of grain with each quickly adjusted matters.

The animals were bred without reference to the experiment as it seems certain from Eckles (6) work that the nutrients required by the fetus could have had no appreciable influence upon the results.

The weights

The plan of the experiment was based upon the conception that an animal kept at uniform weight will use its food only for maintenance and milk production. This plan reduces the factor of gain or loss of weight to the minimum. With the exception of Fusion on late silage and Fruition on early silage, in the first stages, the weights were entirely satisfactory.

The animals were weighed daily. Weighings were made between 9:00 and 10:00 a.m. and the average of the day's weight with the nine preceding days was recorded opposite the daily weight. This ten-day average was charted each day along with the daily weight, and this information provided the basis for grain feeding as described under "feeding the grain."

Type of ration for each group remained the same

The cows were kept on the same type of rations throughout. The rations were not sufficiently different in character to warrant alternating the groups. Such a procedure would also have increased the difficulties of keeping the weight uniform. With the amount of silage and hay the same throughout for each group, (the common practice in feeding) and the weight constant, the only variables would be the amount of milk produced and the amount of grain eaten.

In this plan the basic idea was to have absolute control over the animals, securing accurate results rather than assuming that the variations in each group would offset each other as when working with large numbers.

Preliminary feeding

All animals were placed upon the experimental hay and grain ration, but using the regular herd silage (Eureka) for preliminary observations. After thirty days, on this they were each given the experimental silage for ten days before the experiment proper was started. During this time they were weighed daily. The ten-day average weight was taken on January 25 as the initial weight and the ten-day average on May 5 as the final.

The results

The experiment was run one hundred days at the end of which time one of the silos was empty and the other two practically so. The average daily grain, hay and silage consumed and the protein and dry matter supplied by each is given in the following table.

TABLE 5

Pounds feed, protein and dry matter consumed per day by each group

GROUP	GRAIN			SILAGE			HAY		
	Amount	Protein	Dry matter	Amount	Protein	Dry matter	Amount	Protein	Dry matter
	pounds	pounds	pounds	pounds	pounds	pounds	pounds	pounds	pounds
Early.....	9.9725	1.9382	8.8129	44.977	1.0688	11.2322	4	0.29	3.8308
Medium.....	8.314	1.6158	7.3430	45.739	0.9957	11.5190	4	0.29	3.8308
Late.....	11.604	2.2553	10.2492	45.537	0.7322	9.0806	4	0.29	3.8308

Table 5 shows the average daily silage consumed by each group to be practically the same: Early consuming 44.977 pounds; medium consuming 45.739 pounds; and late consuming 45.537 pounds. The protein and dry matter supplied by the silage is as follows: early 1.0688 pounds protein, 11.2322 pounds dry matter; late 0.7322 pounds protein, 9.0806 pounds dry matter, giving a difference of nearly 0.3 pound of protein and 2.2 pounds dry matter, which is approximately as much protein as was supplied by two pounds of the grain ration and as much total digestible nutrients as was supplied by 3 pounds. The early group consumed an average of 9.9725 pounds of grain, the medium group 8.314 pounds and the late group 11.604 pounds per day.

The actual difference in grain eaten between the early and late groups was 1.63 pounds per day less by the early.

Table 6 shows the average daily milk production for the early group to be 28.286 pounds; the medium, 22.938 pounds; and the late 29.206 pounds revealing a very even production for the two extreme groups. In fat production the extremes are still closer the early group showing 1.0878 pounds and the late group 1.0788 pounds with the medium at 1.0455 pounds. In total solids the extremes are almost identical, showing 3.5769 pounds for the early and 3.5990 for the late with the medium at 3.1578 pounds per day. This table also gives the average weights as follows: early group, 1054.4; medium, 963.3, and late, 1046.8. It shows an average gain of 17.67 pounds for the early, 2.63 pounds for the

TABLE 6
Average daily production and live weights, per cow

GROUP	MILK	FAT	TOTAL SOLIDS	LIVE WEIGHT	
				Average weight	Gain or loss
	pounds	pounds	pounds		
Early	28.286	1.0878	3.5769	1054.4	+17.67
Medium.....	22.938	1.0455	3.1578	963.3	+2.33
Late	29.206	1.0788	3.5990	1046.8	-14.00

medium, and a loss of 14.00 per cent for the late. These changes in weight are extremely small and yet some significance can be attached to them in interpreting the results of the feeding values of the silage.

Table 7 gives the total dry matter, the dry matter per pound of milk, the dry matter per pound of milk solids and the dry matter in grain per pound of milk solids per day by groups. It would appear at first glance that the dry matter in the late silage was more efficient than that of the early and medium silage. However, when it is considered that the group of three on the early silage, each gained 17.67 pounds in one hundred days and that the late silage group of three lost 14 pounds each in the same period, the utilization must be considered about equalized. A striking difference in the dry matter supplied by the grain for each pound

of solids produced in the milk is indicated in the last column of the table.

The relatively high grain requirements shown in table 8 are due to the limited amount of hay fed. It is shown clearly that the milk, fat, and total solids were produced more efficiently by the early silage group than the late group. The good showing of the medium group with regard to milk production and especially in economy of fat and total solid yields is due to their lesser maintenance requirements. The silage analysis would place

TABLE 7

Dry matter consumed and the amount used per unit of production

GROUP	DRY MATTER PER COW PER DAY	DRY MATTER PER POUND MILK	DRY MATTER PER POUND OF SOLIDS	DRY MATTER IN GRAIN PER POUND MILK SOLIDS
	<i>pounds</i>			
Early.....	23.8759	0.8441	6.4794	2.3916
Medium.....	22.7928	0.9937	7.2179	2.3253
Late.....	23.1606	0.7930	6.2962	2.8478

TABLE 8

Production per unit of grain

GROUP	GRAIN PER 100 POUNDS MILK	MILK PER POUND GRAIN	GRAIN PER POUND FAT	GRAIN PER POUND SOLIDS
Early.....	35.405	2.84	8.095	2.752
Medium.....	35.816	2.76	7.923	2.593
Late.....	39.780	2.54	10.838	3.249

this group a close second to the early which is indicated quite effectively by the grain requirement, 35.405 pounds being required by the early group as against 35.816 for the medium, for each 100 pounds of milk. It is striking that every animal in the early and medium groups except one Guernsey in the medium group, produced more efficiently with respect to grain consumed than the members of the late group.

Table 9 represents the values of the early silage by 100 and compares the other types on this basis, using the feeding values in table 8, the yields as determined by the agronomy department

at the Storrs Station and the silage analysis as given in table 3. Here, the medium shows up stronger than the early due, as explained, to the lesser maintenance requirements of the medium group.

The late silage corn yields 167 tons to each 100 tons of the early. In dry matter the relation stands 123 to 100, this ratio being about the same as the fat and solids production per unit of grain, but with the advantage reversed.

TABLE 9
Relation of values—early silage group expressed as unity

GROUP	GRAIN PER 100 POUNDS MILK	MILK PER POUND GRAIN	GRAIN PER POUND FAT	GRAIN PER POUND SOLIDS	YIELD PER ACRE	
					Green weight	Dry matter
Early.....	100	100	100	100	100 (12 tons)	100 (6500 pounds)
Medium.....	101	97	88	94	133 (16 tons)	111 (7200 pounds)
Late.....	112	88	121	118	167 (20 tons)	123 (8000 pounds)

TABLE 10
Grain saved by early and medium silage and value at two cents per pound

GROUP	PER 100 POUNDS MILK		PER 5000 POUNDS MILK	
	Amount	Value	Amount	Value
	<i>pounds</i>		<i>pounds</i>	
Early.....	4 37	\$0.09	219	\$4.38
Medium.....	3 96	0.08	196	3.92

Table 10 shows that a saving 4.37 pounds of grain, having a value of 9 cents was effected per 100 pounds of milk by the early silage. On a basis of 5000 pounds of milk produced this would make a grain saving of \$4.38. The medium group shows a saving of \$3.92 per 5000 pounds of milk.

SUMMARY

1. The experiment is an attempt to determine the relative economy of milk production by growing early, medium or late maturing varieties of corn for silage.

2. Late maturing varieties, under favorable conditions, will decidedly out-yield the early varieties, both in total tonnage and dry matter.

3. The varieties used to represent the three types were: Early (Pride of the North), 25.74 per cent dry matter in silage; medium (Leaming), 25.17 per cent dry matter in silage; late (Eureka), 19.43 dry matter in silage. The season allowed Leaming to mature more than usual, resulting in practically the same per cent of dry matter as Pride of the North and very similar results in the feeding tests.

4. The grain ration consisted of 3 parts corn meal, 3 parts wheat bran and 2 parts cotton seed meal (36 per cent) having a nutritive ratio of 1 to 3.805 and carrying 0.148 pound digestible protein and 0.7171 pound digestible nutrients per pound.

5. The early silage group of cows and the late group each contained two Holsteins and one Jersey and the medium group was comprised of one Jersey, one Guernsey and one Holstein. The object was especially to have the extreme groups evenly balanced throughout.

6. The feeding trial proper covered one hundred days. Thirty days preliminary feeding upon the regular herd silage and ten days upon the experimental silage preceded the experiment proper.

7. Two animals in each group received 50 pounds of silage daily, the third, of less capacity, receiving 40 pounds. The average consumption was 44.977 pounds for the early group, 45.739 for the medium group and 45.537 pounds for the late group. The dry matter furnished by the silage per day to the early group was 11.232 pounds, to the medium, 11.519 pounds, and to the late, 9.081 pounds. The hay consumed was 4 pounds each which was eaten without waste.

8. The grain was fed according to the weight of the animal, the object being to keep them at a uniform weight. By this plan most of the food was used for maintenance and production and a small, unappreciable amount for developing the fetus in early gestation. The average daily amount of grain consumed was 9.9725 pounds by the early group, 8.314 pounds by the medium, and 11.604 pounds by the late.

9. The average daily milk yield was 28.286, 22.938 and 29.206 pounds respectively, by the early, medium and late. The fat yield was 1.0878, 1.0455, and 1.0788 pounds, respectively. The

total solids yield was 3.5769, 3.1578, and 3.5990 pounds respectively.

10. The early group averaged 1054.4 pounds in weight, and gained an average of 17.67 pounds; the medium group averaged 963.3 pounds and gained 2.33 pounds each; the late group averaged 1046.8 pounds and lost an average of 14 pounds.

11. The early group consumed 35.405 pounds of grain per 100 pounds of milk; the medium group consumed 35.816 pounds; and the late group 39.78 pounds. For total solids the early group consumed 2.752 pounds of grain to each pound produced; the medium group 2.593; and the late group 3.249. The better showing made by the medium group in solids production compared to the early is due to their lesser maintenance requirements. The relatively high grain requirement per unit of production for all of the groups is due to the small hay allowance.

12. The results show a saving for 100 pounds of milk of 4.37 pounds of grain for the early group as compared with the late group. Ton for ton, these data seem to indicate greater economy of the early silage. This is further emphasized by the fact that the early group gained some in weight and the late group lost slightly.

The real test comes when the three types of corn are compared on an acre basis, or their efficiency in milk production per acre. The authors do not feel justified in making this comparison until a second feeding trial shall have been completed.

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STUDIES IN THE GROWTH AND NUTRITION OF DAIRY CALVES

IV. THE FEED COST OF GROWING DAIRY HEIFERS

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Every year five to six million heifers are needed to replace the old and unproductive cows that are weeded out of the dairy herds of the United States and to increase the number of cows required to augment the supply of dairy products needed by the country.

Unfortunately in too many cases little attention is given to the proper raising of the young heifers that are ultimately to take their place in the producing herds of the country. Many calves are improperly fed from birth, while in an even larger number of cases the calves are reared well until weaning time when they are turned loose to rustle for themselves. In either case, when the heifers reach producing age they tend to be stunted and are generally undernourished, unthrifty and in poor condition. Such heifers cannot be expected to do their best work when they come into the producing herd. The improper raising of heifers is a factor of great influence in causing the low average production of milk and butterfat in many of the dairy herds of today.

Many men make a practice of raising heifers and then selling them without having an idea as to the cost of production. The feed cost of raising heifers exceeds in amount all the other cost items combined and it was this fact which prompted this study.

RÉSUMÉ OF PREVIOUS WORK

Few trials are reported on the feed cost of raising dairy heifers and only one of those reported carries the heifers through to producing age. In reviewing the literature therefore, only the

studies which carry the heifers to two years of age or over are considered.

Of the three studies reported only the one carrying the heifers to producing age and one of those carrying them to two years of age include large enough numbers of animals to be of the greatest significance.

TABLE 16
Feed required by dairy heifers

	AUTHORITY		
	Bennett and Cooper (11)	Hayden (13)	Trueman (15)
Number of heifers.....	17	37	5
Months fed.....	24	Fresh at 26½	24
Feeds:			
Whole milk, pounds	342	459	445
Skim milk, pounds	3165	3330	2953
Grain, pounds.....	547	1710	737
Dry roughage, pounds.....	2649	2634	3145
Silage and soiling pounds.....	3603	4042	2938
Pasture, days.....	294	322	300

EXPERIMENTAL WORK

The data reported here is obtained from a record of the growth and feed consumption of a group of 40 heifers grown to producing age on the Iowa State College Dairy Farm, in the years 1916 to 1921, inclusive. They all ultimately entered the producing herd.

Purebred and grade heifers of the Holstein, Guernsey and Jersey breeds and purebred Ayrshires are included. No attempt is made to determine breed distinctions as the lots would be much too small for that purpose. An effort was made, however, to determine the difference in feed cost of production of heifers that were dropped in fall and winter on the one hand, and those that were dropped in spring and summer on the other hand. Winter calves were taken to be those dropped between October 1 and March 31, while summer calves were considered to be those dropped between April 1 and September 30.

The calves used in this work were being raised to come into the producing herd and were kept under normal conditions. All were allowed to run with their dams for a few days after birth and were then put on whole milk which was fed for periods longer than is generally considered most economical. Gradually all calves were changed over to skim milk instead of whole milk. The skim milk was also fed for a considerable period. As a consequence it will be found that a large number of the calves were receiving whole milk up to near six months of age and skim milk for another period of about six months. In one case whole milk was fed after six months of age and skim milk after one year of age. This shows up in small amounts in the averages.

TABLE 17
Animals used

BREED	WINTER CALVES		SUMMER CALVES		ALL CALVES	
	Number of animals	Per cent of total	Number of animals	Per cent of total	Number of animals	Per cent of total
Ayrshires.....	4	10.0	2	5.0	6	15.0
Guernseys.....	8	20.0	7	17.5	15	37.5
Holsteins.....	9	22.5	3	7.5	12	30.0
Jerseys.....	3	7.5	4	10.0	7	17.5
Total.....	24	60.0	16	40.0	40	100.0

It will be seen, therefore, that the policy was one of liberal milk feeding and concentrates were also allowed in greater proportions than are generally provided. The feeding of grain was started at about a few weeks of age and was allowed liberally when needed. The same holds true for alfalfa hay. Only relatively small amounts of non-leguminous roughages were allowed and they were fed to the heifers only when no other means of disposal was available.

Pasture was allowed at all possible times as was silage though generally not in large amounts as it was usually not available in large amounts after the producing herd had been allowed for. Soiling was provided in small amounts only on special occasions. Salt and water were provided at free will at all times, with the

exception of the calves receiving milk during winter and they had access to water but twice daily.

In distinguishing between the feeding of the winter and summer calves it may be said that the winter calves were not allowed to pasture until spring, while the summer calves were generally a little younger when allowed to pasture. During their first summer the winter calves were allowed grain liberally and in the fall—when they were a year of age—a more liberal allowance of feed could be allowed them than was given to the summer heifers which were of younger age. In the following summer little grain was allowed in any case, but when the animals of both groups were getting ready to freshen, liberal allowances of grain were given—the winter heifers getting more than the others, as they freshened later in the year.

The practice was to have all heifers, as nearly as possible, freshen in the fall or winter but this was not always possible, due to difference in age. Consequently, it was found on studying both lots, winter- and summer-dropped heifers, that they had come to production at the same average age of twenty-nine months. Throughout this study, calendar months are not used, as it has been found more convenient to consider each monthly period as thirty days; the first period beginning with the day on which the heifer was born.

On consulting the data it is found that the winter calves were 4 pounds heavier than the summer calves at birth but at the age of freshening the winter heifers had made the greater gains and on the average weighed 69 pounds more than the summer heifers at the same age. For convenience the results are combined by periods of six months. It will be found that the winter heifers do not make as good average daily gains in all periods of six months as do the summer heifers but if the average daily gains from birth up to any point be considered it will be found that the winter heifers have always made the best daily gains. At the time of freshening, twenty-nine months of age, the average daily gains from birth were 1.09 pounds for the winter heifers and 1.01 pounds for the summer heifers, or an average for all heifers of 1.05 pounds.

To keep down the size of the paper and yet preserve clarity as far as possible, the amount of concentrates fed per heifer for each period of six months is given for each group and then the total amounts for each heifer are given in cumulative form up to the time of freshening. In addition all the feeds consumed are treated in the same way.

TABLE 18
Average live weight gains per heifer

AGE	WINTER CALVES			SUMMER CALVES			ALL CALVES		
	Weight	Gain for six months	Total gain	Weight	Gain for six months	Total gain	Weight	Gain for six months	Total gain
<i>months</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>
Birth	68			64			67		
6	317	249	249	309	245	245	314	247	247
12	569	252	501	540	231	476	557	243	490
18	750	181	682	698	158	634	729	172	662
24	922	172	854	874	176	810	903	174	836
29	1010	88	942	941	67	877	982	79	915

TABLE 19
Average daily live weight gain per heifer

AGE	WINTER CALVES		SUMMER CALVES		ALL CALVES	
	For six months	For total period	For six months	For total period	For six months	For total months
<i>months</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>
0-6	1.38	1.38	1.36	1.36	1.37	1.37
6-12	1.40	1.39	1.28	1.32	1.35	1.36
12-18	1.05	1.26	0.88	1.19	0.96	1.23
18-24	0.96	1.19	0.98	1.13	0.97	1.16
24-29	0.59	1.09	0.45	1.01	0.53	1.05

The feed prices used were those current at the time of the preparation of the manuscript, November, 1921. The home produced feeds were taken at farm values and did not include cost of marketing. Purchased feeds were taken on the basis of carload lots delivered.

The pasture costs were based on results previously published by Gillette, McCandlish and Kildee (12) which showed that the

average duration of the pasturing season was 167 days in this district. Under the system of management maintained in this work it has been found that one acre of pasture will be sufficient for a heifer of one year old or over for the season. Consequently, by allowing \$8.00 per acre for the pasture, which is a suitable allowance for the rental, manuring and maintenance of the

TABLE 20

Average amount of concentrates consumed by heifers by six-month periods

AGE	CRACKED CORN	HOMINY FEED	GROUND OATS	WHEAT BRAN	OIL MEAL	COTTON-SEED MEAL	TOTAL
Winter calves							
<i>months</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>	<i>pounds</i>
0-6	139	16	79	63	36		333
6-12	468	29	179	154	77		907
12-18	407	18	118	112	59	9	723
18-24	235	2	59	70	48	5	419
24-29	345	4	100	106	88	20	663
Summer calves							
0-6	174	24	82	61	30		371
6-12	396	57	165	131	66		815
12-18	293	25	88	75	39	2	522
18-24	436	88	93	95	51	34	717
24-29	183	8	81	91	73		436
All calves							
0-6	154	19	80	62	34		349
6-12	439	41	174	145	73		872
12-18	361	20	106	97	51	6	641
18-24	314	4	74	82	49	16	541
24-29	280	5	92	99	77	13	566

pasture used, it will be found that the pasture costs for a heifer of twelve months of age or over will be 5 cents per day. It was more difficult to arrive at a cost for the pasture used by younger animals but it is believed that a charge of $2\frac{1}{2}$ cents per day for pasture for animals less than a year of age will be sufficient.

From the prices furnished and the amount of feed consumed by the heifers the cost of concentrates by six-month periods and

TABLE 21
Average cumulative consumption of concentrates by heifers

AGE	CRACKED CORN	HOMINY FEED	GROUND OATS	WHEAT BRAN	OIL MEAL	COTTON-SEED MEAL	TOTAL
Winter calves							
months	pounds	pounds	pounds	pounds	pounds	pounds	pounds
0-6	139	16	79	63	36		333
0-12	607	45	258	217	113		1240
0-18	1014	63	376	329	172	9	1963
0-24	1249	65	435	399	220	14	3382
0-29	1594	69	535	505	308	34	3045
Summer calves							
0-6	174	24	82	61	30		371
0-12	570	81	247	192	96		1186
0-18	863	106	335	267	135	2	1708
0-24	1299	114	428	362	186	36	2425
0-29	1482	122	509	453	259	36	2861
All calves							
0-6	154	19	80	62	34		349
0-12	593	60	254	207	107		1221
0-18	954	80	360	304	158	6	1862
0-24	1270	84	434	386	207	22	2403
0-29	1550	89	526	485	284	35	2969

TABLE 22
Average amount of feed consumed by heifers by six-month periods

AGE	SUCKING	WHOLE MILK	SKIM MILK	CONCENTRATES	SILAGE	SOILING	PASTURE	ALFALFA HAY	CANE FODDER	CORN FODDER
Winter calves										
months	days	pounds	pounds	pounds	pounds	pounds	days	pounds	pounds	pounds
0-6	3.5	1459	712	333	145		21	293		
6-12		57	1386	907	420	16	57	1182	38	21
12-18			7	723	957	51	53	1405	75	152
18-24				419	605	38	130	231	32	501
24-29				663	1959	179	62	463	70	239
Summer calves										
0-6	3.5	1289	954	371	63		25	395		
6-12		26	1229	815	516	18	40	1126		
12-18				522	675		124	353	16	224
18-24				717	1739	16	54	245	202	735
24-29				436	1475	315	125	364	1	123
All calves										
0-6	3.5	1391	809	349	113		22	334		
6-12		45	1323	872	470	17	50	1159	23	13
12-18			3	641	832	31	82	984	52	181
18-24				541	1059	28	100	237	101	594
24-29				566	1765	234	87	425	41	192

TABLE 23
Average cumulative consumption of feed by heifers

AGE	SUCKING	WHOLE MILK	SKIM MILK	CONCENTRATES	SILAGE	SOILING	PASTURE	ALFALFA HAY	CANE FODDER	CORN FODDER
Winter calves										
months	days	pounds	pounds	pounds	pounds	pounds	days	pounds	pounds	pounds
0-6	3 5	1459	712	333	145		21	293		
0-12	3 5	1516	2098	1240	565	16	78	1475	38	21
0-18	3 5	1516	2105	1963	1522	67	131	2880	113	173
0-24	3 5	1516	2105	3382	2127	105	261	3111	145	674
0-29	3 5	1516	2105	3045	4086	284	323	3574	215	913
Summer calves										
0-6	3 5	1289	954	371	63		25	395		
0-12	3 5	1315	2183	1186	579	18	65	1521		
0-18	3 5	1315	2183	1708	1254	18	189	1874	16	224
0-24	3 5	1315	2183	2425	2093	34	243	2119	218	959
0-29	3 5	1315	2183	2861	4468	349	368	2483	219	1082
All calves										
0-6	3 5	1391	809	349	113		22	334		
0-12	3 5	1436	2132	1221	583	17	72	1493	23	13
0-18	3 5	1436	2135	1862	1415	48	154	2477	75	194
0-24	3 5	1436	2135	2403	2474	76	254	2714	176	788
0-29	3 5	1436	2135	2969	4239	310	341	3139	217	980

TABLE 24
Feed prices

	PRICE PER TON
Cracked corn.....	\$10.00
Hominy feed.....	20.00
Ground oats.....	15.00
Wheat bran.....	20.00
Linseed oil meal O. P.....	32.00
Cottonseed meal.....	32.00
Alfalfa hay.....	12.00
Cane fodder.....	8.00
Corn fodder.....	10.00
Corn silage.....	4.50
Soiling.....	4.00
Whole milk, per 100 pounds.....	2.00
Skim milk, per 100 pounds.....	0.25
Sucking, per day.....	0.06
Pasture, per day.....	0.05

the feed cost of production of the heifers, by six-month periods and cumulative from birth to producing age have been prepared.

RESULTS OBTAINED

As has already been noted the heifers used in this study were liberally fed and so the actual feed cost of production will tend

TABLE 25
Average concentrate cost per heifer by six month periods

AGE	CRACKED CORN	HOMINY FEED	GROUND OATS	WHEAT BRAN	OIL MEAL	COTTON-SEED MEAL	TOTAL
Winter calves							
<i>months</i>							
0-6	\$0.70	\$0.16	\$0.59	\$0.63	\$0.58		\$2.66
6-12	2 34	0.29	1.34	1.54	1 23		6.74
12-18	2 04	0 18	0 89	1 12	1.00	\$0.14	5 37
18-24	1.18	0.02	0.44	0.70	0 77	0 08	3 19
24-29	1.73	0 04	0 75	1.06	1 41	0 33	5 32
Summer calves							
0-6	\$0.87	\$0.24	\$0 62	\$0.61	\$0 48		\$2.82
6-12	1 98	0.57	1.24	1 31	1 06		6 16
12-18	1.47	0.25	0 66	0 75	0 62	\$0 03	3 78
18-24	2.18	0 08	0 70	0 95	0 82	0.54	5 27
24-29	0.92	0 08	0 61	0 91	1 17		3.69
All calves							
0-6	\$0 77	\$0 19	\$0 60	\$0.62	\$0 54		\$2.72
6-12	2.20	0 41	1 31	1.45	1.17		6 54
12-18	1.81	0.20	0.80	0 97	0 82	\$0.10	4 70
18-24	1.58	0.04	0.56	0.82	0.78	0 26	4 04
24-29	1.40	0 05	0 69	0 99	1 23	0.21	4 57

to be above rather than below the general average cost of feed required for producing heifers. For the total of 40 heifers the average feed cost of raising them from birth to the time of freshening at the average age of twenty-nine months was \$106.81, while the same figures for the winter and summer heifers were \$109.89 and \$102.43, respectively.

In view of present prices this may seem a high cost of production but it should be remembered that these heifers were fed

liberally and it is liberal feeding that pays. The man who is raising heifers to bring into his own herd cannot afford to stint them on feed as it is only where they have been well grown and liberally fed that the greatest production can be expected from them.

TABLE 26
Average feed cost per heifer by six-month periods

AGE	SUCK- ING	WHOLE MILK	SKIM MILK	CONCEN- TRATES	SILAGE	SOIL- ING	PAS- TURE	ALFAL- FA HAY	CANE FODDER	CORN FODDER	TOTAL COST
Winter calves											
<i>months</i>											
0-6	\$0 21	\$29 18	\$1 78	\$2 66	\$0 33		\$0 53	\$1 76			\$36 45
6-12		1 14	3 47	6 74	0 95	\$0.03	1 43	7.09	\$0 13	\$0 11	21 09
12-18				5 37	2 15	0 10	2 63	8 43	0 30	0 76	19 74
18-24				3 19	1 36	0 08	6 50	1 39	0 13	2.51	15 16
24-29				5 32	4 41	0 36	3.10	2 78	0 28	1.20	17 45
Summer calves											
0-6	\$0 21	\$25 78	\$2 39	\$2 82	\$0 14		\$0 63	\$2 37			\$34 34
6-12		0.52	3 07	6 16	1 16	\$0 04	1 00	6 76			18.71
12-18				3 78	1 52		6 20	2.12	\$0 06	\$1.14	14 82
18-24				5 27	3 91	0 03	2 70	1 47	0 81	3 68	17.87
24-29				3 69	3 32	0 63	6.25	2.18	0 00	0 62	16 69
All calves											
0-6	\$0 21	\$27 82	\$2 02	\$2.72	\$0 25		\$0 55	\$2 00			\$35 57
6-12		0 90	3.31	6.54	1 06	\$0 03	1.25	6 93	\$0 09	\$0.07	20.18
12-18			0 01	4 70	1 87	0 06	4 10	5 90	0.21	0.91	17.76
18-24				4 04	2.38	0 06	5 00	1 42	0 40	2.97	16.27
24-29				4 57	3 97	0.47	4 35	2.55	0 16	0 96	17.03

This was well illustrated in the case of work with scrubs reported by McCandlish, Gillette and Kildee (14). A number of scrubs reared under poor condition had been obtained and put with the purebred herd. Previously these animals had been very poorly fed but when put with the purebred herd at Iowa State College they were given good feed and care. These scrubs can be divided into three age groups, heifers, four-year-olds, and mature cows, according to the age at which they reached the station.

A summary of all the records made by the animals in the three groups, after a correction for age has been made, shows that those coming as mature cows had an average production of 3168.7 pounds of milk and 153.64 pounds of butterfat, while the four-year-olds averaged 3597.7 pounds of milk and 166.36 pounds of butterfat, and the heifers 4036.1 pounds of milk and

TABLE 27
Average cumulative feed cost per heifer

AGE	SUCK- ING	WHOLE MILK	SKIM MILK	CONCEN- TRATES	SILAGE	SOIL- ING	PAS- TURE	ALFAL- FA HAY	CANE FODDER	CORN FODDER	TOTAL COST
Winter calves											
<i>months</i>											
0-6	\$0.21	\$29.18	\$1.78	\$2.66	\$0.32		\$0.53	\$1.76			\$36.45
0-12	0.21	30.32	5.25	9.40	1.28	\$0.03	1.96	8.85	\$0.13	\$0.11	57.54
0-18	0.21	30.32	5.25	14.77	3.43	0.13	4.59	17.28	0.43	0.87	77.28
0-24	0.21	30.32	5.25	17.96	4.79	0.21	11.09	18.67	0.56	3.38	92.44
0-29	0.21	30.32	5.25	23.28	9.20	0.57	14.19	21.45	0.84	4.58	109.89
Summer calves											
0-6	\$0.21	\$25.78	\$2.39	\$2.82	\$0.14		\$0.63	\$2.37			\$34.34
0-12	0.21	26.30	5.46	8.98	1.30	\$0.04	1.63	9.13			53.05
0-18	0.21	26.30	5.46	12.76	2.82	0.04	7.83	11.25	\$0.06	\$1.14	67.87
0-24	0.21	26.30	5.46	18.03	6.73	0.07	10.53	12.72	0.87	4.82	85.74
0-29	0.21	26.30	5.46	21.72	10.05	0.70	16.78	14.90	0.87	5.44	102.43
All calves											
0-6	\$0.21	\$27.82	\$2.02	\$2.72	\$0.25		\$0.55	\$2.00			\$35.57
0-12	0.21	28.72	5.33	9.26	1.31	\$0.03	1.80	8.93	\$0.09	\$0.07	55.75
0-18	0.21	28.72	5.34	13.96	3.18	0.09	5.90	14.83	0.30	0.98	73.51
0-24	0.21	28.72	5.34	18.00	5.56	0.15	10.90	16.25	0.70	3.95	89.78
0-29	0.21	28.72	5.34	22.57	9.53	0.62	15.25	18.80	0.86	4.91	106.81

191.21 pounds of fat. In other words, the cows that had not received good feed until they were mature, were the poorest producers and taking their production as the basis of comparison, it is found that the four-year-olds produced 14 per cent more milk and 8 per cent more fat, while the heifer group produced 27 per cent more milk and 24 per cent more butterfat. This clearly shows that heifers must be well fed from birth to give maximum yields of milk and fat when they reach producing age.

The largest items in the feed cost of raising the heifers were whole milk, concentrates and alfalfa hay, and it was in these three items only that the higher cost was found for winter than for summer heifers, even though the feeds were charged at the same price at all times.

Whole milk is an expensive feed at all times but if heifers are to be raised well, its liberal use is justified in many cases. The winter heifers received more whole milk on the average

TABLE 28

Average feed cost per pound of live weight gain by six-month periods

AGE	WINTER CALVES	SUMMER CALVES	ALL CALVES
<i>months</i>	<i>cents</i>	<i>cents</i>	<i>cents</i>
0-6	14 6	14 0	14.4
6-12	8 5	8 1	8.3
12-18	10 9	9.4	10.3
18-24	8 8	10.2	9.4
24-29	19.8	24.9	21.6

TABLE 29

Average cumulative feed cost per pound of live weight gain

AGE	WINTER CALVES	SUMMER CALVES	ALL CALVES
<i>months</i>	<i>cents</i>	<i>cents</i>	<i>cents</i>
0-6	14.6	14.0	14.4
0-12	11 5	11.1	11.4
0-18	11.4	10.7	11.1
0-24	10.9	10.6	10.7
0-29	11.7	11.7	11.7

than did the summer calves, as they were longer in being turned to pasture. The same facts hold true for the concentrates and the alfalfa hay. Though alfalfa hay is not advocated for calves in this section, yet it was necessary to feed it to all classes of young stock, as it was grown for the milking herd and older heifers and the inclusion of clover hay production in the rotation would have rendered the farming operations more cumbersome.

It has been noted that the feed cost of growing fall- and winter-dropped calves to producing age was greater in this work than was the cost of growing spring and summer calves to producing

age, though the average age of freshening was the same for both groups, twenty-nine months and it is generally conceded that the raising of fall and winter calves is the more profitable proposition. There are certain factors regarding weather conditions and the amount of labor available which do render the raising of fall and winter calves the better proposition but here the feed cost only will be considered.

It will be found by studying the feed cost per pound of live weight gain that for each six-month period, up to eighteen months of age, the feed cost per pound of gain was greater in the case of the winter heifers, but from that age until freshening the feed cost per pound of gain was lower in the case of the winter heifers. Of more importance, however, is the fact that from birth to freshening, at the average of twenty-nine months the average feed cost per pound of live weight gain was the same for each group—11.7 cents.

Consequently, the fall- and winter-dropped calves were the better proposition, as at the age of freshening they had reached a greater weight than the spring and summer born heifers, and had been produced at the same feed cost per pound.

SUMMARY

As a result of the study of the feed cost of growing 40 dairy heifers from birth to freshening, the following statements may be presented for consideration.

1. Of the 40 heifers studied, 24 dropped between October 1 and March 31, were classed as winter heifers, while the remaining 16, dropped between April 1 and September 30, were classed as summer heifers.

2. The average age of freshening in each lot was twenty-nine months; each month being a period of thirty days and not a calendar month.

3. The average birth weights were 68 pounds for the winter heifers, 64 pounds for the summer heifers and 67 pounds for all of the animals.

4. The average weights at freshening were 1010 pounds, 941 pounds and 982 pounds for the winter, summer and all groups

respectively, while the average live weight gains from birth to freshening for these groups were 942 pounds, 877 pounds and 915 pounds.

5. For average daily live weight gains through the trial the winter heifers led with 1.09 pounds, while the summer heifers had 1.01 pounds, and the average for all was 1.05 pounds.

6. In total feed cost of production the ranking was: winter heifers, \$109.89; summer heifers, \$102.43, and all heifers, \$106.81 each.

7. The average feed cost per pound of increase in live weight was 11.7 cents in all groups.

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YEAST AND MOLD COUNTS AND THEIR RELATION TO PASTEURIZATION OF CREAM FOR BUTTER MAKING PURPOSES¹

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I. INTRODUCTION

The yeast and mold content of finished butter is considered by some men² who are engaged in dairy work as an index to the efficiency of pasteurization of cream for butter making purposes and to the proper handling of the cream after this important process is completed. The laboratory of the American Association of Creamery Butter Manufacturers uses the yeast and mold count as an index to efficiency. They assume that when samples of butter show a low count, the product has been manufactured efficiently. From the biological standpoint it has been found that in plants³ producing butter with a high yeast and mold count lax methods of pasteurization and handling of cream prevail. In such plants the counts were lowered materially in the finished products by improving the process of manufacture in regard to the careful pasteurization and handling of the cream.

II. OBJECT

The purpose of this investigation was two-fold: first, to determine whether or not the yeast and mold count could be taken as an efficiency index to pasteurization and handling of cream for butter making purposes; and second, to determine the source of contamination when butter made from pasteurized cream has a high yeast and mold count.

¹ This article was prepared under the direct supervision of Dr. H. A. Ruehe and Dr. M. J. Prucha, Dairy Department, University of Illinois.

² Mr. F. W. Bouska and Mr. J. C. Brown (9) and Mr. T. H. Lund (4, 5, 6, 7, 8).

³ The plants are operated by members of The American Association of Creamery Butter Manufacturers and are located in various communities according to a personal interview with Mr. F. W. Bouska.

III. LITERATURE

There is little published data on yeasts and molds in creamery butter. Mr. T. H. Lund of Ontario Agricultural College, Guelph, Ontario (Canada), has shown that the yeast and mold count in butter is influenced by efficient pasteurization and he has suggested that such a count be adopted officially in determining whether butter is made from raw or from pasteurized cream. Lund has pointed out also, that even though the pasteurization process may be efficient, the cream can be contaminated in the subsequent handling by the use of yeast and mold infected churns or other equipment (5, 6, 8).

Few, if any, yeasts and molds or their spores can survive a temperature exposure of 145°F. for thirty minutes or 185°F. for thirty seconds. Such temperature exposures are those used ordinarily in pasteurizing cream for butter making. Both Dougherty (1) and Wells (2) have demonstrated that most yeasts are destroyed at a temperature of 61°C. (141.8°F.). Dougherty found only three yeasts which survived this temperature. Thom and Ayers (10) have shown that few molds or their spores in milk survive a temperature of 60°C. (140°F.) for thirty minutes and still less are able to withstand 62.8°C. (145°F.) for thirty minutes. Only three species of *Aspergillus* survived 62.8°C. for thirty minutes.

O. F. Hunziker (3) summarizes the efficiency of pasteurization in destroying yeasts and molds in butter. By this investigation, he showed that the efficiency of pasteurization in killing yeasts and molds was 78 per cent when a temperature exposure of 165°F. for thirty seconds was used, while at 185°F. for thirty seconds or 145°F. for thirty minutes (the latter two being the methods ordinarily used in creameries), the process was 99.9 per cent efficient.

Mr. F. W. Bouska has set the arbitrary standard of 30 as the maximum count of yeasts and molds in butter which has been made from efficiently pasteurized cream. However, a large percentage of the counts should be 10 or less (9).

The yeasts and molds, that are found in butter, include a variety of yeasts (not isolated individually) but the molds are mainly *Oidia (lactis)* (9).

IV. METHOD OF PROCEDURE

a. Whey agar

During the first five months of the investigation, whey agar was used as a medium for determining the yeast and mold counts. The whey agar was prepared as follows: 15 grams of shredded agar and 10 grams of "Difco" standardized bacto-peptone in 1 litre of whey. The whey was secured by adding 1 cc. of rennet extract (diluted to 40 cc. with cold water) to a gallon of skimmed milk, and after the coagulation was completed, the curd was filtered off through a double thickness of cheese cloth.

Approximately 10 cc. of the above medium was used for each petri dish, and to this 1 cc. of a sterile 1 per cent tartaric acid solution was added for the purpose of inhibiting the growth of all organisms other than yeasts and molds. However, it is possible that some *Bulgaricus* bacteria may grow on such medium.

b. "Malt" agar

Later in the investigation, a medium made from ordinary malt drinks (often sold under the name of "near beer") was found to be more satisfactory for yeast and mold growth than the whey agar. The "malt" agar was made as follows: 15 grams of shredded agar was added to 400 cc. of the malt drink and 600 cc. of distilled water. This was heated to boiling and held at this temperature until the agar was dissolved. The medium was acidified just previous to pouring the plates by mixing 4 cc. of a sterile 5 per cent solution of lactic acid with 100 cc. of "malt" agar. This acidity was sufficient to inhibit the growth of organisms other than yeasts and molds. This medium was similar to that used by Mr. T. H. Lund, but varied in one of its ingredients. The agar prepared by Mr. Lund contained brewery wort (8), while in this investigation, brewery wort was not obtainable and the malt drink was substituted for it.

To obtain as complete data as possible, counts were run on butters which were made from both pasteurized and raw cream. In order to determine yeast and mold counts, every step in the butter making process was plated. In the case of the butter made from unpasteurized cream, counts were made on raw cream and butter. In the case of pasteurized cream, plates were made from raw cream, pasteurized cream, ripened cream, butter, buttermilk and starter. In all cases, except the butter, 1 cc. of the sample was plated by the ordinary procedure for using petri dishes. In the case of the butter, it was melted to the constituency of thick cream and 1 cc. was used for each count. The most rapid and luxurious growth of yeasts and molds was obtained by incubating the plates at 30°C. for a period of five days.

The sample of the raw cream was taken from the vat just previous to pasteurization. After the entire volume of cream was pasteurized, the cream in the vat was agitated and a sample was taken for plating. After pasteurization, the cream was cooled to 70°F. and 10 per cent starter was added. It was held at this temperature for one and one half to two hours and then the cream was cooled to the churning temperature and held over night. A sample of this ripened cream was taken just previous to churning. When the churning was completed and the butter worked, a representative sample (some butter from various parts of the churn) was taken and the plates were made. The sample of buttermilk was drawn from the gate of the churn immediately after the butter granules had gathered. In the case of the starter, the sample was taken after the starter was mixed thoroughly by stirring, and previous to its addition to the pasteurized cream. All of the above liquid samples were taken by using a sterile 100-cc. pipette. A sterile spoon was used for sampling the butter. The samples were placed in sterile ground-glass-stoppered bottles of 250 cc. capacity.

For the purpose of checking up on yeast and mold infected churns, the following procedure was carried out with three large churns (600 and 1000 pounds capacity) of the type used in commercial creameries and one small power churn (75 pounds capacity). In the case of the large churns, 20 gallons of boiled

water were put in the churn and it was revolved in fast gear for five minutes. The same method was used in rinsing the small churn except that 5 gallons of boiled water were used. In each case, a sample of the boiled water was plated to obtain a count, so that the increase in the microorganism content of the water after rinsing could be taken as the increase due to yeast and mold infected churns.

Plates were made from samples of starter secured from various creameries in the State of Illinois to determine the influence of average starters as a source of contamination in pasteurized cream. Yeast and mold determinations were made on each sample.

V. DISCUSSION OF DATA

As the results in table 1 indicate, the yeast and mold content may be reduced to a small number in the cream by pasteurization, whereas the butter may show an increase. This tends to show that the yeast and mold content can not be taken as an index for efficiency in pasteurization of raw cream. The increase of yeasts and mold in butter over that in the pasteurized cream is due, no doubt to the contamination in the subsequent handling of the cream.

Although the yeast and mold content in butter cannot be taken as an efficiency index to pasteurization, the data points to the fact that it can be considered as an efficiency index to the entire butter making process. This is true if we adopt a maximum count of 30 (Mr. F. W. Bouska's standard) yeast and mold colonies as efficient. In table 1, there are only five samples of butter out of a total of twenty-five with a higher count than 30. Three of the five contained a small number of yeasts and molds above the maximum or 30. When the maximum is passed, the cause of the high count can be traced to inferior starter or to unsterile apparatus, especially to yeast and mold infected churns.

The results indicate that a large percentage of the counts in butter show a decrease over the pasteurized cream, yet the ripened cream carries a higher count than either. The buttermilk shows a decided increase over the above counts, and this tends to in-

dicates that a large number of the yeasts and molds are not incorporated in the butter but are washed out in the buttermilk.

The flash method of pasteurization (where a temperature exposure of 185°F. for thirty seconds is used) was found to be 99.9 per cent efficient in killing yeasts and molds. This compares favorably with the results obtained by Hunziker (3). The percentage of the decrease from the raw cream to the finished butter is found in table 2. The percentage of decrease in yeasts and

TABLE 1
Yeast and mold counts in pasteurized cream butter

COUNTS OF YEASTS AND MOLDS PER CUBIC CENTIMETER	NUMBER OF SAMPLES					
	Raw cream	Pasteur- ized cream	Ripened cream	Butter	Buttermilk	Starter
0	0	0	0	1	0	5
1-5	0	9	2	4	0	2
6-10	0	3	1	4	4	0
11-30	0	8	8	11	2	2
31-50	0	1	2	3	4	1
51-100	0	1	2	1	1	0
101-500	0	2	0	1	0	0
501-1,000	0	0	1	0	1	0
1,001-5,000	2	0	1	0	1	3
5,001-10,000	0	0	0	0	0	0
10,001-50,000	6	0	0	0	0	0
50,001-75,000	3	0	0	0	0	0
75,001-100,000	1	0	0	0	0	0
100,001-150,000	4	0	0	0	0	0
Above 150,000	9	0	0	0	0	0

molds is smaller, as a rule, than it is after pasteurization. This points directly to the fact that the cream may be efficiently pasteurized and later recontaminated.

It has been suggested by Mr. Lund that the yeast and mold counts be taken as a method for distinguishing pasteurized from unpasteurized cream butter. The results in table 1 show that cream may be efficiently pasteurized but later recontaminated. In comparing table 1 with table 3, it is shown that yeast and mold counts cannot be used effectively. Table 1 shows that cream which has been pasteurized may carry a high count in the butter.

This may be due to inefficient pasteurization but the main cause is recontamination in the subsequent handling. In table 3, it is shown that unpasteurized cream butter had a lower count of yeasts and molds than in some cases of pasteurized cream butter.

TABLE 2
Yeast and mold reduction

PERCENTAGE RANGE	NUMBER OF SAMPLES	
	Pasteurization efficiency	Decrease from raw cream to butter
99.9 and over	11	12
99.95-99.98	7	7
99.90-99.94	5	2
99.80-99.89	2	0
99.50-99.79	0	2
99.00-99.49	0	0
Under 99.00	0	2

TABLE 3
Yeast and mold counts in unpasteurized cream butter

COUNTS OF YEASTS AND MOLDS PER CUBIC CENTIMETER	YEASTS AND MOLDS	
	Raw cream	Butter
0-75	0	0
76-150	0	1
151-500	0	2
501-1,000	2	0
1,001-5,000	1	1
5,001-10,000	0	2
10,001-25,000	1	1
25,001-35,000	0	1
35,001-50,000	3	0
Above, 50,000	1	0

There is no doubt but that the churn is a source of contamination for the finished butter. It is a difficult task to rid the churn of yeasts and molds by ordinary methods, i.e. using hot water followed by steam. The churn will harbor these organisms in the pores of the wood, boxings, cracks and similar places where treatment is almost impossible.

From table 1, it can be seen that the churn is a source of contamination. The total yeast and mold count in the butter and in the buttermilk is greater than the count of the ripened cream previous to being run into the churn. This difference must be due to the churn either through direct contamination or the breaking up of clumps by agitation. During the churning process, the agitation is sufficient to break clumps into several parts, and this is no doubt a factor in causing the large increase in yeast and mold counts in the butter and buttermilk over that in the ripened cream.

TABLE 4

Yeast and mold counts in churn rinsings

COUNTS OF YEASTS AND MOLDS PER CUBIC CENTIMETER	NUMBER OF SAMPLES OF YEASTS		NUMBER OF SAMPLES OF MOLDS		NUMBER OF SAMPLES OF INCREASE DUE TO CHURN	
	Check on water	Number in rinse water	Check on water	Number in rinse water	Yeasts	Molds
0	12	1	10	0	1	1
1-5	2	4	4	6	4	5
6-10	0	2	0	0	2	0
11-50	0	3	0	3	3	3
51-100	0	0	0	0	0	0
101-500	0	1	0	3	1	3
501-5000	0	2	0	1	2	1
Above 5000	0	1	0	1	1	1

In table 4, the results show the effect of the churn in yeast and mold contamination. In studying this table, it must be remembered that in this investigation 20 gallons of rinse water were used in a churn in which ordinarily 200 gallons of cream were churned. Therefore if the rinse water shows a count of 20 yeast and molds per cubic centimeter it would mean the addition of only 1 yeast and mold colony per cubic centimeter if 200 gallons of cream were used.

The ripened cream shows an increase of yeasts and molds over the pasteurized cream (table 1). This may have been due to any one or all of four causes: (1) vat contamination; (2) breaking up of colonies by agitation in the vat; (3) increase due to growth or multiplication; and (4) the starter used. Steaming the vat for thirty minutes apparently killed all of the organisms. At least

plates made from water which had been sterilized and then used to rinse the vat did not show a yeast or mold colony. The agitation in the vat is slow and probably has little effect in breaking up clumps of yeasts and molds. Counts made upon cream before and after agitation confirmed this opinion. After pasteurization was completed and the starter was added, the cream was cooled to 45°F. to 50°F. (depending on the season of the year) and held over night. The tendency of such a low temperature would be to inhibit multiplication of all organisms.

At first, the starter was thought to be the main source of contamination. However, the average starter is low in yeast and mold content (table 5) and plays a rather unimportant part when

TABLE 5
Yeast and mold counts in starters

COUNTS OF YEASTS AND MOLDS PER CUBIC CENTIMETER	NUMBER OF SAMPLES	
	Yeasts	Molds
0	17	11
1-3	6	12
4-7	2	3
8-12	1	0
13-18	2	1
19-25	0	1
Above 25	0	0

we consider that ordinarily only 10 per cent starter (20 gallons in 200 gallons of ripened cream) was used in the plant where this investigation was carried out. For example, if 20 gallons of starter containing 20 yeast and mold colonies per cubic centimeter are diluted to 200 gallons with cream, it would mean an addition of one colony per cubic centimeter in the cream. None of the starters plated (table 5) contained as many as 20 yeast and mold colonies per cubic centimeter and eleven contained none. This tends to show that the starter may be considered a negligible factor in contamination except in rare cases when an inferior grade of starter is used. In table 1 there are four examples of highly contaminated starter, but the butter in two cases out of the four shown will come within the standard of 30 yeast and mold colonies per cubic centimeter of butter.

VI. CONCLUSIONS

1. The yeast and mold count of finished butter cannot be taken as an efficiency index to pasteurization. However, if a standard of 30 colonies is set as the maximum count of these in the finished butter, it can be considered as an efficiency index to the entire butter making process.

2. The data point to the fact that the yeast and mold count cannot be taken as an effective method for determining whether butter is made from raw or from pasteurized cream.

3. The churn may be one of the greatest sources of contamination of the cream after pasteurization.

4. The cream may be contaminated when starter is used, but this source of yeasts and molds has little effect on the final count in the butter except in rare cases when an extremely inferior grade of starter is added to the cream.

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LABORATORY SUPERVISION OF PASTEURIZATION PLANTS

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Inspection of the dairies will undoubtedly reduce the danger of disease germs getting into milk, but the sole practical method of assurance that it shall contain no disease germs is by adopting pasteurization. The committee on milk supply of the American Public Health Association has concluded (1920) (1) that any change in the chemical composition caused by pasteurization, if there be any, is very small and that any ill effects from the use of such milk may be remedied by the addition of such common substances as orange juice and potato water. "The protection from communicable diseases that pasteurization affords older children and adults far overshadows any of the easily remedied ill effects associated with infant feeding." This committee's findings further show that of approximately 4200 pasteurization plants in the United States and Dominion of Canada, only a very limited number are controlled from a public health point of view. It has been found that pasteurization alone is insufficient in solving the safe milk problem, but that the process must be controlled by high grade supervision and frequent inspection. It is also essential that systematic inspection of producing farms be employed. Pasteurization is not intended to cover up the trail of dirty milk.

In order to insure reliable results from a pasteurization plant, it is essential that the plant be operated by men who are thoroughly familiar with all the machinery and with the process itself. It is also advisable that the machinery be inspected each day by the manager or one of his assistants, before the milk is started from the raw tank, to make certain that the pipes, vats, and other parts of the equipment have been properly cleaned. This procedure is a matter of routine in some of our modern plants. Another factor which should be given frequent attention is the

TABLE 1

Bacterial counts and percentage reductions from three pasteurization plants, by months, July, 1919 to August, 1920

MONTH	NUMBER OF SERIES OF SAMPLES	PLANT	BACTERIA PER CUBIC CENTIMETER					
			Raw	Clarified	Pasteurization tube	Reduction per cent	Bottles	Reduction per cent
July... ..	2	1	12, 000, 000	1, 000, 000	40, 000	92.8	2, 000, 000	83.3
		2						
		3						
August.....	21	1	125, 866, 000	230, 000, 000	1, 485, 000	98 0	7, 181, 000	94 2
		2						
		3	150, 600, 000				150, 000	99 8
September..	15	1	5, 746, 000	5, 813, 000	142, 000	97.8	416, 000	
		2						
		3	4, 404, 000				3, 000	99.9
October. ...	7	1	1, 788, 000	1, 880, 000	7, 000	99 9	8, 000	99 9
		2						
		3	140, 000				1, 000	99.2
November..	3	1	536, 000	563, 000	4, 000	99 2	27, 000	93 0
		2	3, 400, 000		22, 000	99 3	27, 000	99.1
		3						
December..	5	1	760, 000	2, 425, 000	3, 000	99 5	10, 000	90 8
		2	3, 025, 000		39, 000	97 0	65, 000	97.8
January....	2	1	1, 200, 000	Clarification discontinued.	2, 000	98 3	2, 000	98 3
		2	402, 000		66, 000	83 5	94, 000	76.6
February..	3	1	830, 000		2, 000	99.7	5, 000	99.3
		2	516, 000		62, 000	87 9	51, 000	90.1
March.....	2	1	900, 000		13, 000	98.5	14, 000	98.5
		2	633, 000		17, 000	97.3	9, 000	98.5
April.....	2	1	2, 100, 000		8, 000	99.6	4, 000	99.8
		2	1, 100, 000		2, 000	99 8	8, 000	99.2
May.....	1	1	1, 000, 000		2, 000	99.8	2, 000	99.8
		2	800, 000		500, 000	37.5	400, 000	50.0
June.....	1	1	1, 000, 000		2, 000	99.8	2, 000	99.8
		2	1, 900, 000		400, 000	78.9	400, 000	78.9
July.....	3	1	3, 600, 000		3, 000	99.9	10, 000	99.4
		2	2, 200, 000		8, 000	99.6	20, 000	98.6
August.....	2	1	990, 000		2, 000	99.7	2, 000	99.7
		2	230, 000		2, 000	99.1	2, 000	99.1

constant use and accuracy of recording thermometers. It is not sufficient to obtain a high reduction in number of bacteria; the milk must be submitted to a temperature sufficient to kill disease organisms and must be protected thereafter from recontamination. The mere fact that milk passes through a pasteurization plant is not a sufficient guarantee of safety, for improperly cleaned cans, bottles, or careless methods may lead to trouble. The findings of the author from three pasteurization plants in a southern city of about 37,000 population further illustrate the importance of pasteurization-plant inspection and supervision.

The above figures indicate that the bacterial counts for raw milk for all plants were higher in the first months of this study. The milk pasteurized in plant no. 1 for the most part, came from dealers within a radius of 20 miles. The sampling in the plant was made from the raw-mixing tank and was a composite of all the milk. Special attention to the cleaning of this tank, together with prompt cooling of the milk, resulted in lower counts from the raw tank thereafter. As would be expected, the clarified milk gave a higher count, in practically every instance, than the raw milk before passage through this machine, due in large measure, undoubtedly, to the breaking up of "clumps," which would otherwise develop into single colonies. There is also the possibility of inoculation from an improperly sterilized machine.

The pasteurization results from all plants showed the pasteurizers to be effective most of the time except in the case of plant no. 2, a coöperative creamery which received shipped milk from different parts of the state. The fact that they had no recording thermometer, coupled with the use of old machinery by careless helpers was undoubtedly responsible for the erratic results noted. After closing the plant in July, 1920, a general clean-up was instituted and additional equipment was added. Under new management the plant reopened a short time later and the succeeding results were satisfactory.

In case of dealer no. 3, whose dairy farm nearby was the source of supply, the early raw milk counts were excessive in numbers for a modern milk plant. Greater care in cleaning the milking machine, with the use of chloride of lime, according to suggestions

of Taylor of the Dairy Division, U. S. Department of Agriculture (2) together with an increase in period of sterilization of cans, produced striking results in lowering the raw milk count. The low counts recorded in September and October were maintained for the rest of the year. Recontamination of the milk after pasteurization is believed to account for the frequent increase in bacterial counts after bottling, for none of these plants pasteurized in the bottle. It is possible to obtain a noticeable difference in counts of the same lot of milk plated at the same time, as has been conclusively demonstrated in the extensive New York studies (3) supervised by the late Professor Conn. It is believed, however, that the high counts in the bottled milk of this study are the direct result of inoculation from the bottling machine, or the bottles or other sources operating after the milk left the heating tank. This idea was confirmed by special studies of equipment (4) which indicated in plant no. 1, from studies made in July and August, 1919, a possible contamination of milk from the bottling machine of from 2500 to 9000 bacteria per cubic centimeter if the machine were full, and a possible inoculation from bottles of from 20 to 600 bacteria per cubic centimeter. Results from plant no. 2 were even higher from a study of several series of six tests for each series.

The process of can sterilization was also studied and revealed interesting results. It was found that the usual time given to can sterilization was about ten seconds of exposure to live steam, and the rinse water tested from these cans showed a possible inoculation from both cans and covers of 1000 bacteria per cubic centimeter. Ordinarily, these cans received no further treatment by the dairymen before the addition of new milk.

No special study was made of bottle caps, but Smith (5) clearly demonstrated in his study of pasteurization and subsequent handling of milk that while the initial inoculation from milk bottle caps may be small, the importance of handling and of storage of the caps, especially in bulk lots, should not be overlooked.

Following our study of the mechanical features in plant no. 1, new apparatus for sterilization of equipment was installed

and careful supervision of the process instituted. This resulted in the production of a good quality of "pasteurized milk" which increased in popularity.

CONCLUSIONS

The owners of creameries and milk depots want maximum efficiency from their plants. Lack of information concerning methods or lack of supervision of important details may prevent the attainment of best results. Health department supervision should be combined with plant supervision in order that public and plant may be protected. Coöperation results in profit for both the public and the plant.

Contamination of the milk after pasteurization is a very important problem, and careful supervision should be exercised. Medical inspection of the employees in a milk plant should be carried on, and all those coming in contact with the milk after pasteurization, in the bottling, capping, etc., should be given prophylactic inoculation where this is possible. Reinfection of milk injures the results of pasteurization and may be the source of spreading disease. Pasteurization does not necessarily mean protection. Pasteurization, coupled with intelligent and consistent dairy inspection at the source, and competent supervision at the milk plant will insure safe milk.

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A NOTE ON THE WEINZIRL ANAEROBIC SPORE TEST FOR DETERMINING MANURIAL POLLUTION OF MILK

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The public is slowly being trained to demand clean milk. It is becoming more and more the function of the laboratory investigator to apply methods for determining this and from time to time to devise new methods. Among the several different methods which have recently been proposed for this purpose is the so-called anaerobic spore test as proposed by Weinzirl (1) who claimed that the test was superior to either estimations of visible dirt and enumeration of bacteria and that the simplicity of the test along with the simple apparatus required, made it very easily applied. The test according to Weinzirl was carried out as follows:

One-half to 1 cc. of melted paraffin is placed in a 15-mm. test tube which is plugged with cotton and sterilized either by dry or moist heat. By means of a sterile pipette, 5 cc. of milk under test are then placed in the Arnold sterilizer and heated to 80°C. for from ten to fifteen minutes. This treatment melts the paraffin which rises to the surface where it hardens on cooling and forms the anaerobic seal. The heat also expels oxygen absorbed by milk thus rendering anaerobiosis more complete. All the vegetative bacteria are killed by the heat only the spore forms remaining. The tubes are then incubated for three days at 37°C. If anaerobes are present, gas will be formed which will lift the paraffin plug in the tube. Two positive out of five tubes shows excessive pollution.

While the literature on this test is not voluminous, several investigations have been reported. The only one that need be mentioned in this note is the publication of Ayers and Clemmer (2) who have given a summary of literature and report of an investigation on the same subject. They stated, "The Weinzirl test does not appear to correlate with the quantity of manure in

milk. This seems to be due both to the method of making the test and to the variation in spore content of *B. enteritidis* sporogenes in manure." The investigation here reported was carried out before the paper by Ayers and Clemmer was seen.

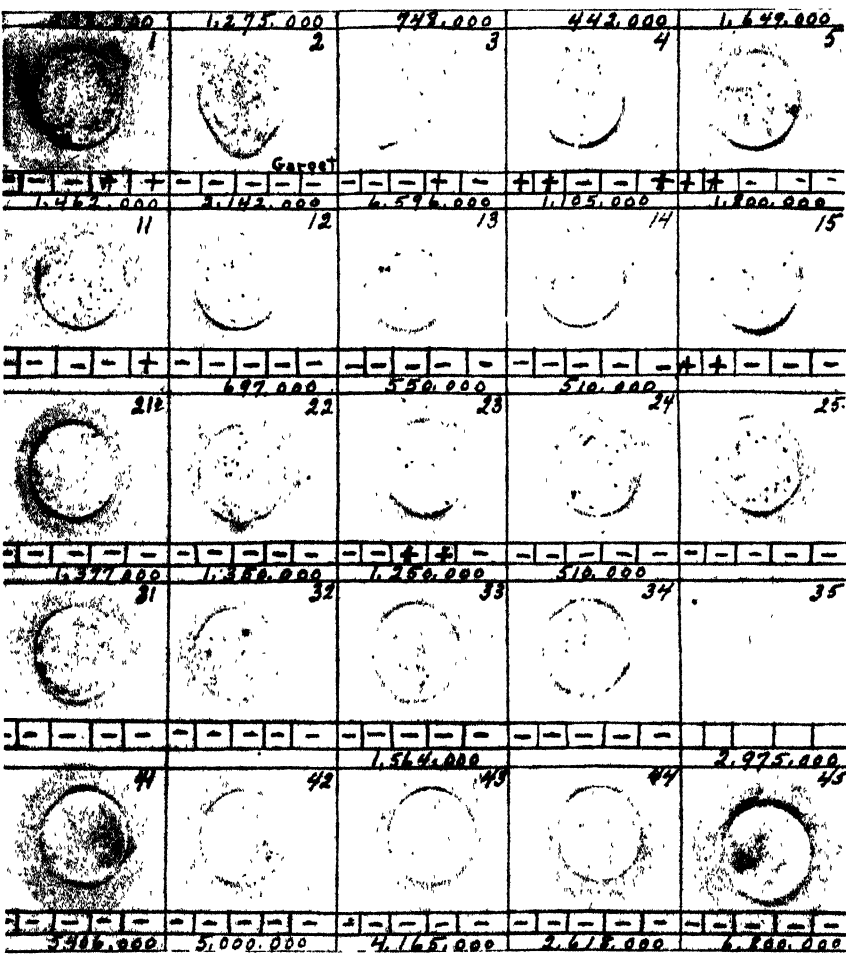


FIG. 1

In order to determine the validity of this test under practical working conditions the authors carried out an examination of over one hundred samples of milk taken at the Illinois Dairy Company at Springfield, Illinois. The samples were collected early in the spring of 1921 over a period of several days. They

were collected in 1-pint sterile bottles and sealed with sterile pasteboard caps. Ten of these samples were taken at one time and stored in the cooler at about 40°F. until they were taken to the laboratory for examination. Twenty-five samples were taken each day on four consecutive days with absolutely no at-

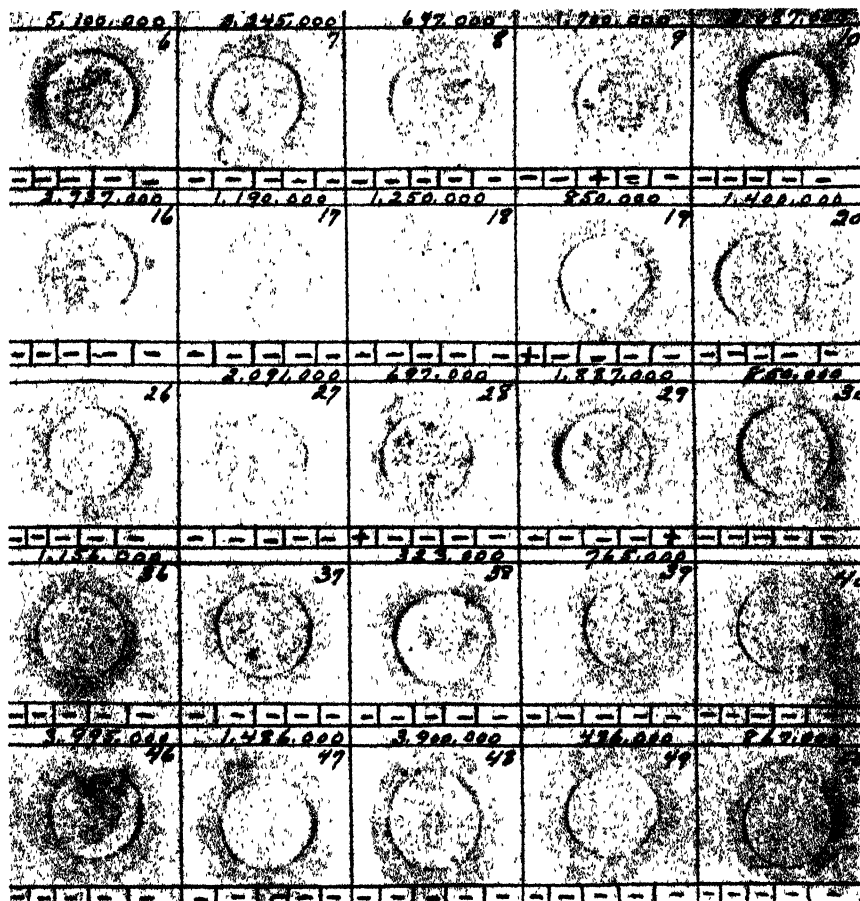


FIG. 2

tempt at selection. In order to avoid any possibility of artificial selection, milk was taken from the first twenty-five patrons which came to the creamery each day. On account of the character of the experiment we used the Frost plate count and made sediment determinations with a Wizard sediment tester since these two determinations may be taken as fair indexes of milk quality.

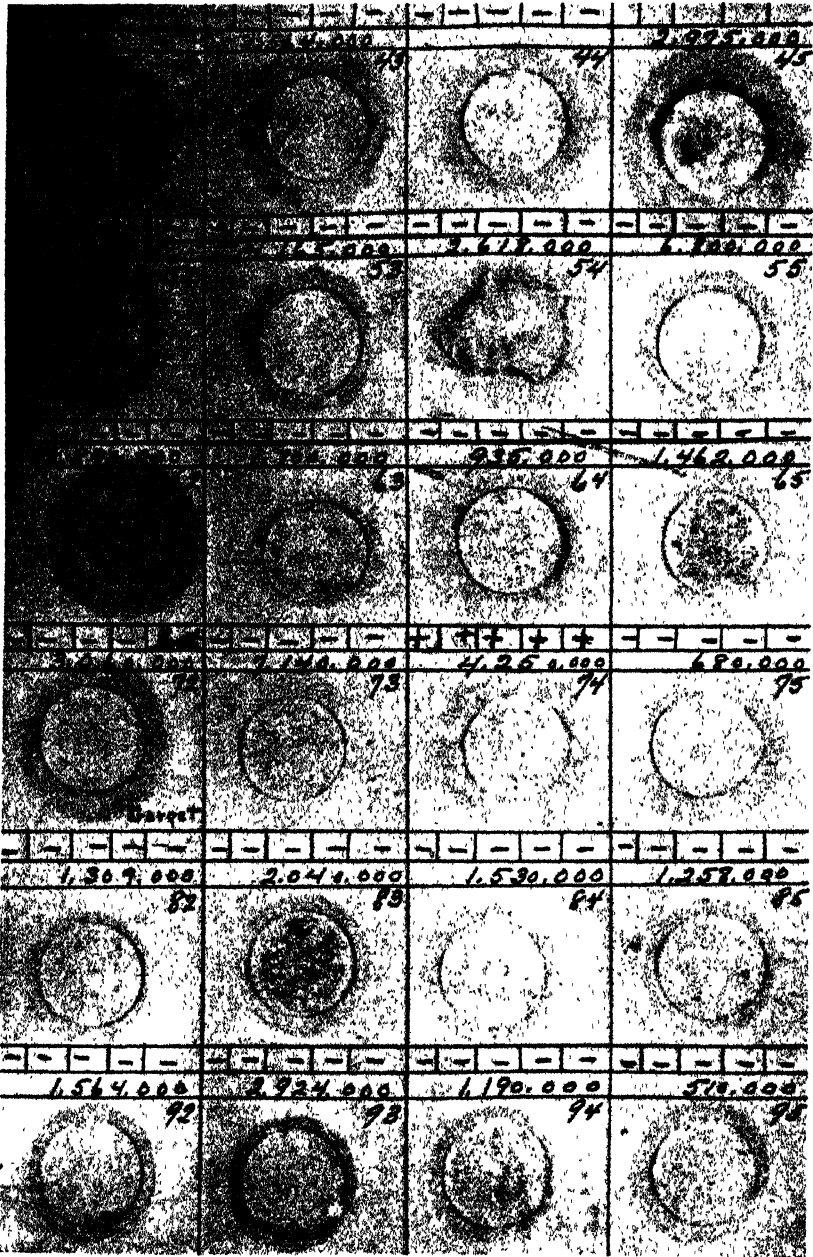


FIG. 3

The spore test which we are studying in this examination was made exactly in accordance with the directions quoted above from Weinzirl's communication. The bacterial count in the spore test sample was taken from the milk before it was passed through the sediment tester.

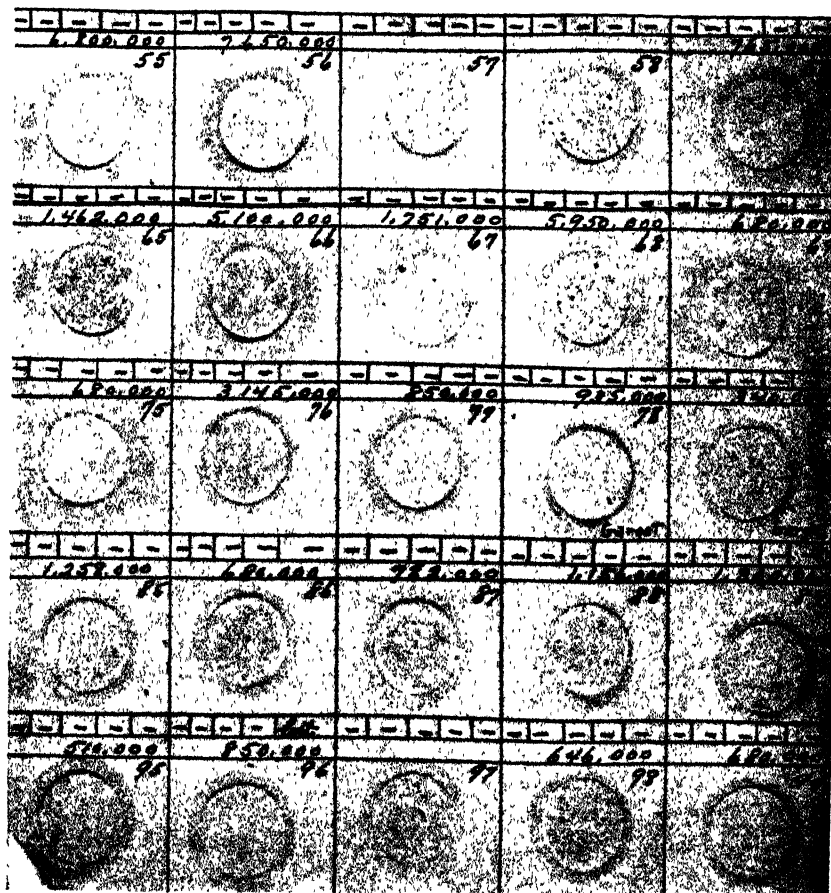


FIG. 4

Some of the results of the analyses are indicated on the illustrations accompanying this paper. An examination of these data shows that there is no correlation between the anaerobic spore test as outlined by Weinzirl and the amount of dirt in the milk, or the number of bacteria as determined by the Frost microscopic plate. For instance, sample 74 had 4,250,000 but

was negative for anaerobic spore forming bacteria. Another illustration of this is sample 46. Others could be mentioned. According to Weinzirl two positive out of five tubes used in the spore test indicated excessive pollution. In our work many samples had two or more positives but very low numbers of bacteria and very small amounts of dirt.

SUMMARY AND CONCLUSIONS

From this investigation it seems that the anaerobic spore test as outlined by Weinzirl cannot be accepted as a method for accurately determining manurial pollution in milk. In this investigation there seems to have been no correlation between the amount of dirt and the number of bacteria, nor the results from the spore test as outlined by Weinzirl. In this way our results confirmed those reported by Ayers and Clemmer.

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THE RELATIONSHIP BETWEEN THE HYDROGEN-ION CONCENTRATION AND THE BACTERIAL CONTENT OF COMMERCIAL MILK

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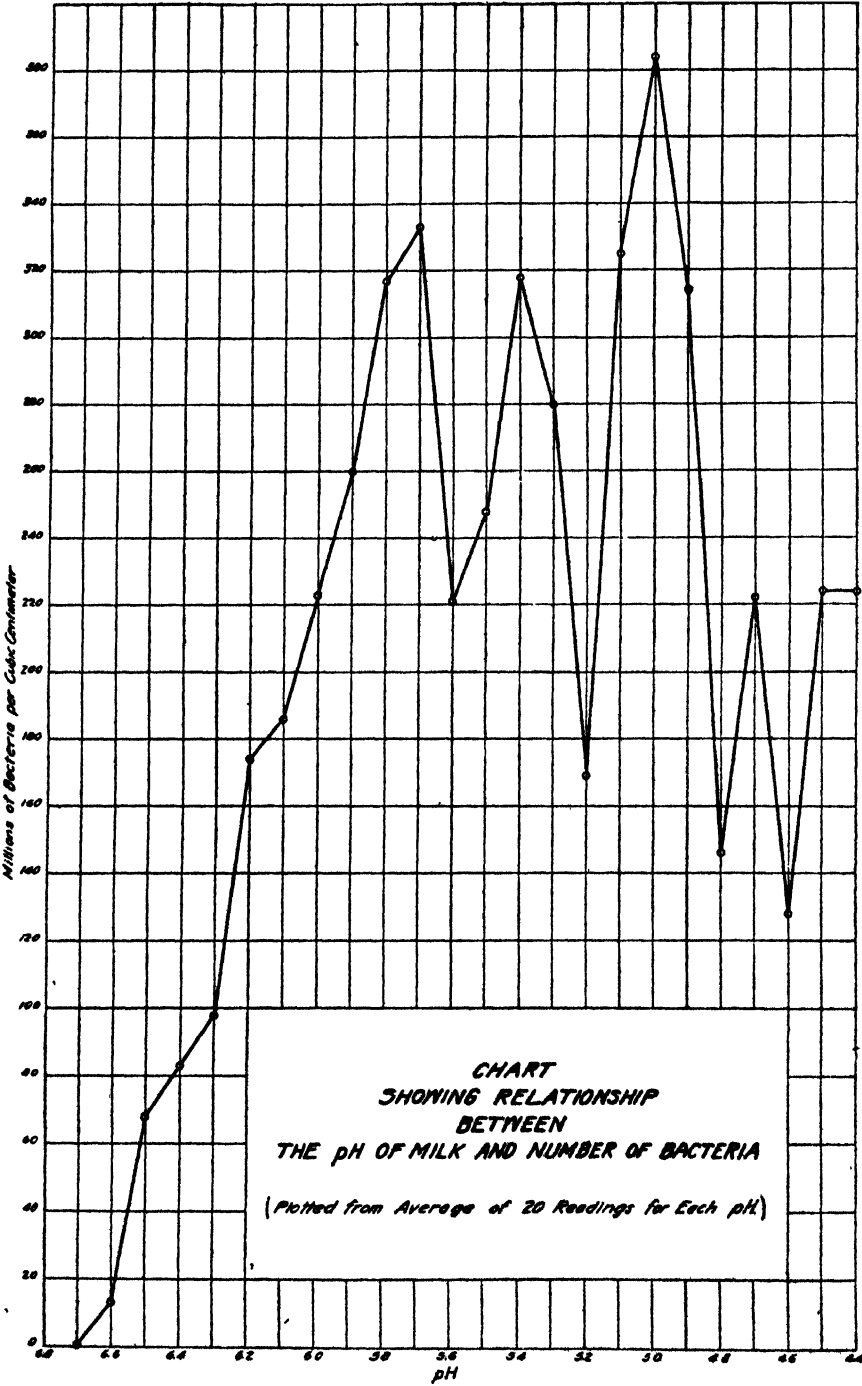
These studies were undertaken, as indicated in our preliminary report (1), with the hope that out of them would eventually grow a practical, indirect, method of ascertaining in a few minutes the probable number of bacteria in commercial milk. Making a bacterial count of milk by the standard plate method is a matter of forty-eight hours, while the hydrogen-ion concentration may be determined by the colorimetric method in less than ten minutes. It was hoped that an indirect method of this kind would not only serve to estimate the number of bacteria in market milk at any given time, but would also indicate its probable age and keeping qualities.

Instead of the colorimetric method described in the previous paper most of the subsequent hydrogen-ion determinations were made with a Wendt's electro-titration apparatus. No appreciable difference, however, was noted when the two methods were checked against each other. The colorimetric method yields itself far more readily to universal employment, which is to be considered in the practical application of this method of determining the number of bacteria in market milk, should further studies warrant its adoption.

These determinations were made on pooled milk obtained from two city dairies and one dairy farm. The bacterial counts were made according to the standard set by the American Public Health Association in 1916. The milk was plated immediately after making the pH readings.

RESULTS

The figures tabulated below represent the averages and the extremes in bacterial count of twenty unchosen determinations. That is, when the goal of twenty determinations for a given pH



was reached no further studies were made on milk at that pH, and so on until twenty determinations were made for each pH from 6.8 to 4.4.

The chart was prepared from the average counts given in the tabulation. It is, we believe, self-explanatory.

TABLE I
Summary of results of twenty determinations for each pH

pH	HIGHEST COUNT	LOWEST COUNT	AVERAGE COUNT
6.8	110,000	750	27,500
6.7	2,500,000	15,000	395,000
6.6	68,000,000	200,000	13,000,000
6.5	325,000,000	2,500,000	68,000,000
6.4	275,000,000	5,000,000	83,000,000
6.3	300,000,000	500,000	98,000,000
6.2	2,000,000,000	2,000,000	174,000,000
6.1	700,000,000	2,000,000	186,000,000
6.0	800,000,000	15,000,000	223,000,000
5.9	1,000,000,000	10,000,000	260,000,000
5.8	600,000,000	5,000,000	317,000,000
5.7	800,000,000	16,000,000	333,000,000
5.6	500,000,000	6,000,000	221,000,000
5.5	600,000,000	7,500,000	248,000,000
5.4	700,000,000	66,000,000	318,000,000
5.3	800,000,000	4,000,000	280,000,000
5.2	500,000,000	10,000,000	169,000,000
5.1	900,000,000	5,000,000	325,000,000
5.0	1,500,000,000	6,000,000	384,000,000
4.9	800,000,000	3,000,000	314,000,000
4.8	400,000,000	2,000,000	146,000,000
4.7	600,000,000	4,000,000	222,000,000
4.6	800,000,000	2,000,000	128,000,000
4.5	500,000,000	40,000,000	224,000,000
4.4	400,000,000	64,000,000	224,000,000

DISCUSSION

Our figures show that with slight increases in hydrogen ion concentration may go enormous increases in the bacterial count. For example, fresh milk usually gave a pH reading of about 6.8 or 6.7; milk in which acidity is just detectable by taste, a reading of

about 6.0. This difference of 0.8 in the pH reading corresponds to a difference in the average bacterial counts of more than 222,000,000, or 27,000,000 for each tenth of a division in the pH scale. Furthermore, a deviation of only a tenth or two from the normal pH is associated with a bacterial count which exceeds the legal limits of most cities. Even the lowest count at 6.6 exceeds the maximum allowed in many places, certainly the average count of 13,000,000 at this pH does. At 6.5 the bacterial counts do definitely exceed the numbers generally permitted though the change in acidity cannot be detected by taste. Not only can one say that the bacterial content is exceeded but that in consequence of this the keeping quality of the milk is seriously impaired.

This brings us to an explanation of the rather marked differences between the lowest and highest count at the various pH readings. They are less disconcerting when studied together with the average counts, representing in most instances rare extremes in the count. Still, in addition to divergences which may be simply an outgrowth of technique, there must be kept in mind the fact that the flora of milk is not constant and that bacteria vary among themselves in their ferment-producing activities. Cooledge and Wyant (2) found that while the bacterial count corresponded in a general way with the keeping quality of milk, that the rate of fermentation as determined by the colorimetric method was a more accurate index.

The reason for the oscillations in the average counts from 5.7 to 4.4 is not clear. It is possibly related to environmental changes—the hydrogen-ion concentration influencing the flora in one way or another. At any rate, the curve is of practical significance only from 6.6 to about 5.8, where the changes in the milk are readily enough detected.

CONCLUSIONS

The results of our studies indicate that the quality of market milk may be estimated with reasonable accuracy by measuring its hydrogen-ion concentration. Since the latter can be deter-

mined in a simple and rapid manner colorimetrically, this means of judging the quality of milk should find a large field of usefulness in the dairy industry and in the hands of consumers, especially in hospitals, etc. where it is desirable to know the quality of milk before its consumption.

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THE PHYSICAL ANALYSIS OF DRY MILK

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With the increased appreciation of the place of the dairy cow as a converter of roughage and other waste into human food, and with the very great appreciation developed within the past few years of milk as a food, the attention of the scientific, the medical, the dairy, and the engineering worlds has been focused closely upon the dehydration of milk, more especially of whole milk. This was done as a means of overcoming the two great handicaps of the realization of better agriculture and better nutrition, namely, the bulkiness and the perishability of milk.

The problems in the manufacture of a standard high class, clean, and nutritious powdered skim milk have been very satisfactorily solved. The problem, however, which until recently most seriously confronted the dairy industry and the human feeding problems of the present time was the production of a powdered whole milk so made that the fat would remain sweet, non-tallowy, and non-rancid for a time sufficiently long to permit of normal marketing methods, with little loss from spoilage. For two-thirds of a century men have fought this problem with definite progress being recorded about every ten to fifteen years, until at the present time a good full-fat milk in powder form may be obtained, with sufficient effort, in most sections of the civilized world.

There are, of course, problems yet to be solved, and these, in my judgment, can be studied by what may be termed physical analysis to better advantage than by chemical analysis alone. While the latter will indicate to us the nominal food value of the product it gives us little or no idea of the structure of the powder particle, nor of the condition or position of the fatty portion within the powder, and yet upon these two things hinge, to a considerable extent, the keeping quality of all dehydrated whole

milks. The various physical aspects of the product may be briefly considered as follows:

COLOR

The color of powdered milk is but a poor guide as to its fat content for the reason that the color will vary with the feed of the cows, as with liquid milk and cream. The amount of heat applied or the cooking given also influences the color. The higher temperature or longer time produces a deeper hue, though of a brownish, rather than a yellow tint. The coarser grained powder also has a richer appearance, due to the greater absorption of the light rays in the coarser product, and its shattered reflection in the finer powder.

TEXTURE

The dehydrated milk possessing smooth, silky feel rather than the coarse, harsh condition is generally more desirable, not alone for the reason merely of the sensation produced, but due in greater measure to experience that coarse milk is usually difficult to put into solution. The finer the powder grain the more easily it will go into solution, up to about 75 microns in diameter, beyond which greater fineness is undesirable, since the particles of such powder dissolve on the outside of little lumps, and form a paste which protects the other fine particles within, and render solution less readily accomplished than would be the case with a slightly coarser grained powder, one which will allow the water to penetrate the mass. An average of about 150 microns in diameter seems most desirable.

ODOR

It is said that "the nose knows," but in the case of milk the nose may know too much. Not infrequently dehydrated milk of various types will give off an unpleasant odor when the tin is first opened, but will still reconstitute into a liquid milk wholly above criticism. Desiccated whole milk is far more likely to become tallowy than rancid or in other words, oxida-

tion rather than hydrolization takes place. But in the case of either the nose can detect its presence or its approach in advance of any but the most extremely delicate chemical test, and far ahead of the tongue.

TASTE

Powder which may reveal the first traces of oxidation of fat to the nose or which may give some of the so-called "condensed milk taste," when tasted in the powder form will not, when constituted and cooled, give any concern. Moreover, when all the people learn that the "cooked" taste is a sweet taste, made by the processes that protect them, they will look for and even seek that "sweet taste of safety."

SOLUBILITY

To meet the needs of the home trade the dehydrated whole milk should be very readily soluble in water of moderate temperature; that is, a temperature of 80 to 120°F. or not warmer than the hand could readily bear. Good powder reconstituted in such warm or semi-hot water should go into essentially complete solution in from fifteen seconds to half a minute, though the writer has repeatedly performed this task on a home scale in half of such length of time. Any milk requiring scalding temperature to dissolve the product has but limited place in the home, and would also raise the question as to whether some alkali had not been used. By solubility in this connection the writer has in mind what may be termed practical solubility; that is the dissolving of the particle to such point that no undissolved specks or flakes will appear on the glass; no portion settle to the bottom upon standing, and which will give a practically normal reading for milk in the cryoscope when reconstituted to a liquid milk having 12.50 per cent to 12.75 per cent total solids—3.5 of which are fat. Powdered whole milk is now being made, which regularly gives such satisfactory results without the addition of any alkali to facilitate solution. Any dehydrated milk which after reconstitution will, upon standing, separate showing a heavy sediment in the bottom of the glass,

and a watery zone above, is not suitably soluble even though it may have a merky, milky look when freshly stirred.

CREAM RISING

At last a system of dehydration of whole milk has been developed which will permit true cream to rise when the powder is reconstituted. Not merely a little cream or little mass of melted butter fat, but a goodly amount of true cream, practically like that on any well pastuerized liquid milk. This fete was accomplished by the development of the Dick, or Economic System, which system uses no vacuum pan, pressure spray nor drum.

It is difficult to convince the housewife that there is cream in any milk unless she can see it. Thus the struggle to develop pasteurizing machines and methods which shall render ordinary market milk safe and which will still permit the cream to rise. Moreover, unless the cream will rise on reconstituted milk it cannot be made available for use in coffee or on cereals as generally desired. The power of cream to rise or rather the power of fat to rise and drag with it portions of the milk plasma may be said to be the difference in the specific gravity between the fat and the surrounding plasma multiplied by the mass of the fat, less the retarding effect of the more or less vicid surrounding plasma. Consequently, any treatment which increases the viscosity of the plasma or sub-divides the fat into minute particles retards or prevents the cream rising by reducing the upward effect of the fat until it is less than the strength of viscosity in the plasma, retarding the fat. Such prevention of creaming is one of the reasons for the development of the homogenizer and its use in the ice cream industry. For the fat to rise as a true cream the butter fat globule must not have been melted as in the case of the vacuum pan, and to rise at all it must not have been broken up by the force of pressure spraying.

DOUBLE COLORING

It is helpful in the testing of the cream rising quality of any milk whether raw, pasteurized, or reconstituted, to add a little Soudan III solution, or other fat soluble stain. It emphasizes

the cream line. Then by adding some water-soluble stain of another color, for instance, hematoxylin, or better common green ink, the green body of the skim milk may be made to stand out in striking contrast to the orange-red of the cream; or if the cream has not risen, the location of the fat in the mass of milk is readily seen. Some milks when reconstituted show a decided sediment of undissolved milk when allowed to stand a

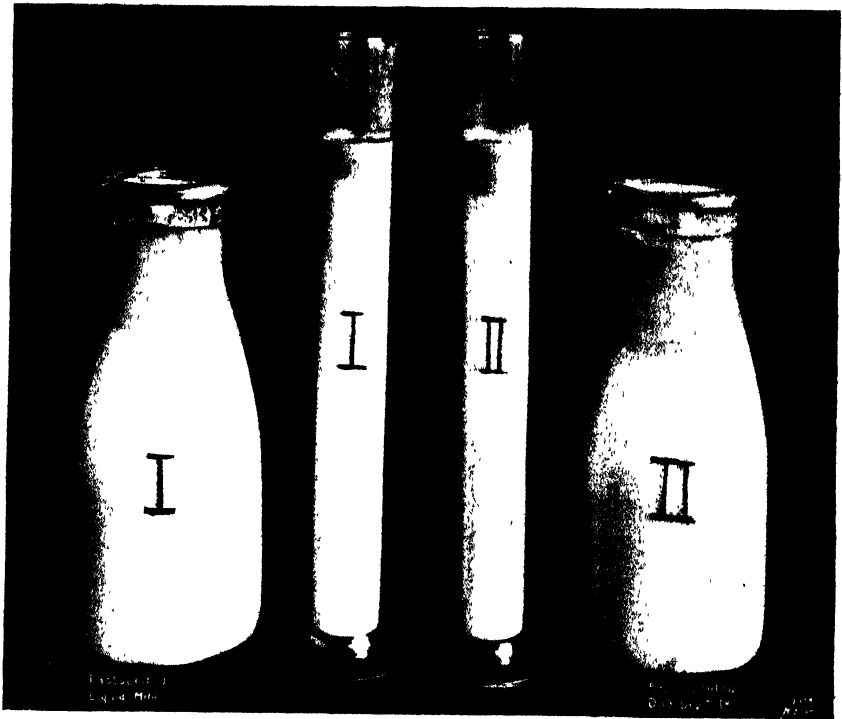


FIG. 1

few hours. Double coloring brings out these and other features. In the study of the cream rising qualities of reconstituted milk of the type most recently developed compared with normal city milk, as shown in this article, figure 1, Soudan III was used. This explains the dark color of the cream here shown. The liquid or natural milk, no. 1, is pasteurized and contains 3.45 per cent fat while no. 2 is reconstituted powdered pasteurized milk, containing 3.60 per cent fat.

CREAM SEPARATION

One test for the condition of the fat globule in any given dry milk is to reconstitute it and run same through an ordinary cream separator. If cream may be separated from the reconstituted milk, leaving only the nominal trace of fat in the skimmed milk and producing a cream which may be churned into normal butter in the ordinary way, it may be assumed that the butter fat globule has not been materially broken up by the process of manufacturing the powdered, or otherwise dehydrated milk.

DIFFERENTIAL STAINING OF POWDER GRAINS

Probably the most interesting single feature in the physical analysis of dehydrated milk is the examination of the entire, unbroken, undissolved milk grains or fragments under the microscope, especially after they have been stained, and more particularly, after the double or differential staining has been applied. "If the process employed (in the production of desiccated milk) is such as to destroy the globular form of the fat globules, it is impossible to reduce the dried milk to a homogeneous fluid, similar to normal fresh milk" (Hunziker). And the form or condition of the fat particle can be studied by staining it a bright orange without separating it from the non-fatty constituents in which it is imbedded, and better yet, when these non-fatty elements are stained a bright blue. Thus doubly stained the condition of the fat particles and their location within the powder grains, especially with respect to their exposure to the atmosphere may be clearly studied.

The sensitiveness of most of the spray process milk powders toward oxidizing agents is augmented by the fact the atomizing process under high pressure causes a subdivision of the fat globules, depriving the fat of at least a portion of the protective gelatinous layer which surrounds each original fat globule, thereby exposing more directly to the destructive oxidizing agents.

Obviously, other things being equal, that full cream milk will keep best in which the fat globules have not been either melted

together as in the condensing pan, nor yet broken into small bids, as in the pressure spray system of dehydration. Dr. Storch showed, to his own satisfaction, many years ago that each and every fat globule was surrounded by a minute but definite gelatinous envelope, which protected the fat from oxidation, so long as it was allowed to remain unbroken. The writer has not been able as yet to stain this envelope of the fat separately from the other non-fatty elements of the powder grains, but he has succeeded in staining the fat one color, and the non-fatty portion of the powder grain another color, and this, too, without dissolving or destroying the original form of the powder grain itself. This was done by the use of the fat soluble reddish stain Soudan III followed by the water soluble blue hematoxylin, after having fixed or hardened the powder grains by brief application of ethyl alcohol 65 per cent strength. The only really difficult portion of the whole task of differential staining is in so fixing the powder that it will not be dissolved by later applications of either water or alcohol, which of necessity must form a considerable portion of stains as vehicle for the color itself. The method of staining the fat globules and their location with respect to the non-fatty portion of the milk particles, which the writer has found best adapted to his needs, and which may doubtless serve as basis for similar study by others is as follows:

1. Clean glass slides thoroughly.
2. Rub Mayer's glycerin albumin (1) evenly and not too thickly over portion of surface of slide.
3. Touch or scatter small amount of powder on the slide not too thickly so that particles do not overlap one another. Single layer deep is best.
4. Treat with ethyl alcohol 65 per cent strength one-half to one minute to harden the proteins of the powder to prevent solution later.
5. Apply Soudan III solution (2) two to five minutes, or until desired color is obtained.
6. Treat with 65 per cent ethyl alcohol one-half minute.
7. Wash gently with tap water.
8. Apply hematoxylin solution (3) three to six minutes. This stain varies greatly in strength with age.

9. Wash gently with tap water.
 10. Wipe excess water from around preparation.
 11. Mount in Farrant's gum mountant (4) and cover with coverglass.
- The mount will keep six months to a year if stored in darkness. The blue tends to fade with time.

Reference to stains

1. Mayer's glycerin albumin mixture. Equal parts of egg albumin and glycerin thoroughly beaten and filtered. Add 1 per cent sodium salicylate to prevent decomposition.
 2. Soudan III fat stain, seventy per cent ethyl alcohol supersaturated with Soudan III with the aid of heat. Let cool; then filter and it is ready for use.
 3. Harris hematoxylin. Hematoxylin cryst, 1 gram; ethyl alcohol, 99 per cent, 10 cc.; dissolve hematoxylin in alcohol; alum (ammonium or potassium), 20 grams; distilled water, 200 cc. Bring mixture to a boil as rapidly as possible; then add 0.5 gram mercuric oxid and cool quickly. Filter before using.
 4. Farrant's gum mountant. Picked gum arabic, 60 grams; water, 60 cc., glycerin, 30 cc.
- Keep in stoppered bottle with a lump of camphor.

STUDYING THE STAINED POWDER GRAINS

A microscopic study of many of the various dehydrated milks on the market reveals the presence of essentially only three types: (1) That produced by the film or hot roller process, which gives us under the microscope a ragged looking particle with waves and wrinkles in the blue body of the milk, with reddish fat in comparatively large fragments scattered promiscuously throughout the mass, many being on the very surface, and therefore exposed to the atmosphere; (2) a powder particle made by the vacuum pan and pressure spray system which is fairly uniform in size, inclined to be spherical in form with the fat present in extremely minute particles scattered so uniformly throughout the entire mass as to cause the powder particles to assume a pink or purple color under the microscope instead of showing a field of blue containing bright orange-red spots of fat; and (3) a powder made without precondensing, and without

the pressure spray, which system produces powder grains, quite uniform in size, almost perfectly spherical in form and in which the fat globules remain unbroken, unfused, and so imbedded in the non-fatty portion, and so coated round about by a colloidal covering that it is protected thereby from the oxygen of the atmosphere.

The three types are shown in the illustrations (fig. 2), nos. 1 and 4 being the low and high magnification of the film, hot roller

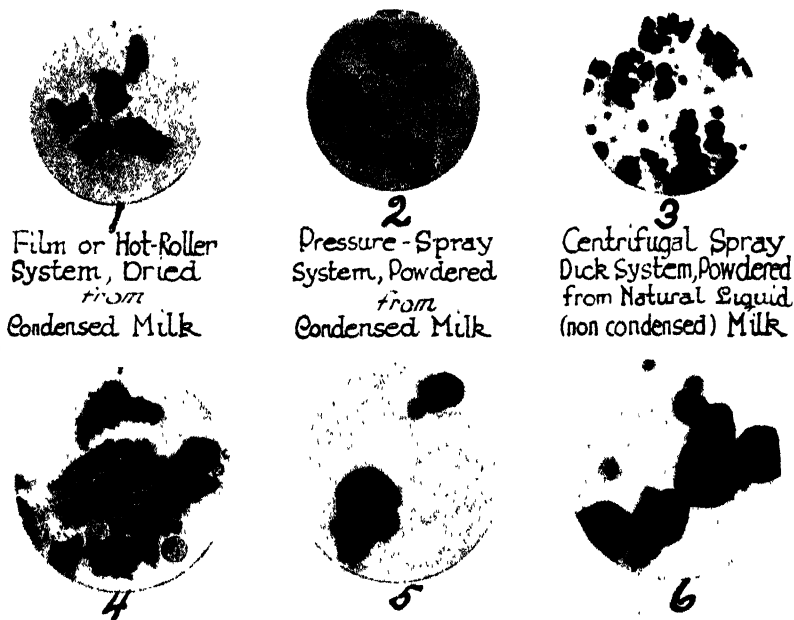


FIG. 2

or drum product. Nos. 2 and 5 are of the pressure sprayed product from condensed milk, and nos. 3 and 6 illustrate the physical makeup of the powder resulting from the Dick process of dehydration. The presence of essentially unbroken globules of fat is indicated by the irregular dark areas (orange-red photographs black) within the powder grain while the blue colloid which surrounds the fat and protects it is shown in the photograph as a light zone (blue takes light). When examining the prepared slide under the micro-

scope by focusing on the smaller powder grains at the point of greatest diameter the enveloping or incysting colloid will be seen as a blue line. The fact that the membrane surrounding the fat has remained unbroken and that the globules are imbedded within the powder grain, surrounded by what is essentially a dry capsule undoubtedly accounts in considerable measure for the unusual keeping quality of powder of this type.

Screwing the microscope down until it crushes a few powder grains, and releases the fat globules, is of passing interest in the study of the physical makeup of powder grains.

The microscope also reveals in the differentially stained powder grain occasional droplets of water, and not infrequently particles of air also within the grain. These are indicated by their refraction.

COLORED LIGHTS

Another interesting phase of this study is the examination of these products unstained but by the aid of lights of different color. Characters which are not discernable with white light become readily seen by means of orange, red or green lights of various intensities. Light intensifiers, as well as the desired colors, may be made by the use of a globular shaped 500-cc. flask filled with water colored with water soluble analine dyes, potassium bichromate, sodium hydroxide and phenolphthalein, or even red and green ink using a common white electric light back of the flask.

SOME PHYSICAL ASPECTS OF DIGESTION

Although milk is generally ingested a liquid, it remains so normally only eight to twelve minutes, by which time the pepsin-rennet ferments, acting in coöperation with the acids of both the milk and the stomach, bring about a curdling. This curd mass begins almost at once to contract and squeeze out the whey, which, carrying most of the sugar and albumen, trickles on into the small intestine where digestion takes place. The mass of curd made up of a mesh of casein filaments, retaining the fat mechanically, remains in the stomach for a considerable

time, three hours or longer, being gradually broken up into small bits and doled out slowly for further digestive action. The retardation seems to form a real step in the best handling of the food.

Reconstituted milks in general respond to this curd formation nearly if not quite as quickly as natural liquid milk, but produce a slightly more tender, even flocculent first curd. After the whey portion has been forced out the remaining curd, if of raw milk, is firm, close in texture and almost rubbery, but if from reconstituted milk is mellow, tender and friable. Curds from reconstituted milks therefore are far more nearly like those goat's and breast milk. They require little or no barley or oatmeal water to cause friability as in the case of natural liquid cows' milk when fed to infants.

The breaking up of the fat globules, in some of the processes, does no harm to the feeding value nor does such improve it. (Confer pages 56, 57, 68, 69, 103, 104, and illustrations opposite pages 32 and 64, Bulletin 195, Vermont Experimental Station, 1916.) From the physical standpoint reconstituted milk is superior to natural raw milk for infant and child feeding.

Fascinating and productive studies may be carried on to the advantage of the dairy industry and all concerned by differential coloring and staining of liquid and dry milk, and by yet closer study into other physical properties and conditions of milk and its products.

A METHOD FOR THE DETERMINATION OF AMINO NITROGEN AND AMMONIA IN CREAM AND BUTTER¹

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It is the general opinion of dairy chemists that the nitrogenous constituents of milk and cream undergo certain changes which begin at an early stage, the degree of change depending, principally, upon the kind and amount of contamination and the temperature at which the product is held. These changes may be caused by the action of enzymes and bacteria upon the proteins with the production of intermediary products of more or less complexity, the simplest being the amino acids. The more favorable the conditions for enzymic and bacterial activity, the greater should be the number of amino groups formed from the original protein. A determination of the amino nitrogen reacting with nitrous acid in a solution from which the undecomposed proteins have been removed by filtration, will be a measure of the proteolysis that has taken place except for the nitrogen that has been synthesized into protein by the microorganisms present. In studying the decomposition of cream and butter, this method is used to show the relation of amino nitrogen to the quality of the product. In the manufacture of butter, some of the nitrogenous constituents of the cream are occluded by the fat and remain in the finished product. In order to measure the relative proportion of amino nitrogen in milk, cream, buttermilk and butter, it is desirable to have a procedure that is adaptable to all of these products. Acetic acid, as used to precipitate the casein in milk will not always work satisfactorily on cream and butter, especially when considerable proteolysis has taken place. In some cases the precipitation of the casein is incomplete and filtration extremely slow and difficult. In the procedure outlined below, picric acid and acetic together have been used satisfactorily to

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separate the protein and higher complex substances from the lower degradation products in milk, cream, buttermilk and butter. Picric acid not only acts as a protein precipitant, but also as a preservative, so that the filtrate can be held for some time without any appreciable change taking place in the amount of nitrogen reacting with nitrous acid. This makes it possible to prepare samples of cream or butter at any creamery so that the analysis can be made later at a fully equipped laboratory.

The determination was made as follows: A 15-gram sample of cream was weighed in a flask of about 100 cc. capacity, 5 cc. of 10 per cent acetic acid and 30 cc. of a saturated solution of picric acid added, and the mixture shaken for half an hour or at intervals for two hours. The solution was filtered on a dry filter and 10 cc. of the filtrate taken for determination of reacting amino nitrogen in Van Slyke's apparatus, using the large reacting bulb and the 10 cc. gas burette. The weight of sample represented by the 10 cc. aliquot of the filtrate, is determined by taking, as the entire volume of solution, the calculated water in the sample of cream and in the added reagents. All results were figured to per cent of the total nitrogen in the sample. For the determination of amino nitrogen in butter, a 100-gram sample was weighed in a 250 cc. ground glass stoppered bottle while the sample was of such a consistency that it could be poured and yet not have any separation of the fat and curd. The bottle was then filled to the shoulder with petroleum ether, boiling point above 55°C., which had been warmed a few degrees above room temperature. The fat was dissolved by shaking the tightly stoppered bottle vigorously for a few seconds and the separation completed by centrifuging and syphoning off the solution, leaving the curd in a compact mass at the bottom. To this residue were added 5 cc. of 10 per cent acetic acid and 30 cc. of a saturated solution of picric acid, the mixture shaken, filtered, and amino nitrogen determined as on the filtrate from cream.

In order to show how completely amino acids could be determined by this procedure, a sample of pure glutamic acid and a portion of the products formed by the acid hydrolysis of casein were added to a sample of butter. The reacting amino nitrogen

was determined as above and the results, as given in table 1, showed that practically all the added amino nitrogen was recovered.

In table 2 are given the results by the above procedure on two kinds of cream. A number of samples were prepared from each cream and the amino nitrogen determined in the filtrates when freshly prepared and after standing the number of days indicated in column 1. Cream no. 1 was a sample of raw cream which had been kept in the ice box forty-nine days, and no. 2 was a sample of fresh sweet cream. The results, as given in columns 2 and 4, show that there was no change in the amount of reacting nitrogen when the filtrate was kept at room temperature for two

TABLE 1

Showing recovery of known amounts of amino acids in filtrate from picric acid and acetic acid precipitation of curd of butter

NUMBER	AMINO ACID ADDED	AMINO NITROGEN		RECOVERY per cent
		Added	Found	
		per cent	per cent	
1	None	None	0 0009	
2	Acid hydrolysis products	0.0751	0 0737	96.9
3	Acid hydrolysis products	0.0738	0.0752	100.7
4	Acid hydrolysis products	0 0369	0.0369	97.6
5	None	None	0.0003	
6	Glutamic acid	0.0188	0.0191	100.0

weeks. To another set of samples acetic acid and picric acid were added and the mixture shaken and kept at room temperature, the time being shown in column 1. These samples were filtered immediately before the determination of amino nitrogen was made. The results, as given in columns 3 and 5, show that there was a slight, though noticeable, increase, in the amount of amino nitrogen.

Table 3 shows the contrast that may be obtained by this procedure on sweet cream and on decomposed cream. In the sweet cream, the amino nitrogen was approximately 2 per cent of the total nitrogen. This figure was increased to 5.8 per cent when the cream was kept at room temperature for six days, and to 19.4 per cent of the total nitrogen after twelve days. Since a sample

TABLE 2
Amino nitrogen as per cent of total nitrogen

DAYS HELD	OLD CREAM		SWEET CREAM	
	Filtered at once	Held without filtering	Filtered at once	Held without filtering
0	{ 6.84 6.76	6.84 6.76	1.81 1.61	1.81 1.61
1	{ 6.58 7.14		2.00 2.15	
2	{ 7.08 7.08	7.11 7.38	1.60 1.59	
3				{ 1.94 1.94
4	{ 6.58 6.76	7.06 7.00	2.08 2.04	
5				{ 2.24 2.22
7			2.07	
8				{ 2.48 2.26
9	6.92 {	7.62 7.62	1.86 1.90	2.66 2.36
10				{ 2.67 2.67
15		{ 7.56 7.77	1.96	
16	6.74			{ 3.20 3.05

of cream from the same source as the above showed only 6.8 per cent amino nitrogen, after being held in the ice box for forty-nine days, it is evident that temperature has a very great influence in the formation of free amino groups from the protein complexes. A similar contrast is shown on some samples of experimental

TABLE 3

Showing amino nitrogen as per cent of total nitrogen in cream samples

NUMBER	DESCRIPTION OF SAMPLES	AMINO NITROGEN AS PER CENT OF TOTAL NITROGEN
1	Sweet cream.....	1.7
2	Sweet cream....	1.7
3	Sweet cream.....	2.2
4	Raw cream held six days at room temperature.....	5 8
5	Above sample held twelve days at room temperature.	19 4
6	Raw cream held in ice box forty-nine days.....	6 8
7	Raw cream held in ice box forty-eight days.....	7 4

butters as shown by the results reported in table 4, the per cent of amino nitrogen in the last sample being approximately 13 times that of sample no. 1, which was a fresh butter from sweet cream.

TABLE 4

Showing amino nitrogen as per cent of total nitrogen in samples of butter varying in quality

NUMBER	DESCRIPTION OF SAMPLE	AMINO NITROGEN IN PER CENT OF TOTAL NITROGEN
1	Fresh butter from sweet cream.....	1.4
2	Fresh butter from neutralized sour cream.....	3.3
3	Experimental butter from neutralized sour cream, held in cold storage six months, then at room tem- perature one week.....	6.5
4	Above sample no. 3, after standing at room tempera- ture one month and twenty days.....	18.1

In order to show the effect upon the amounts of amino nitrogen found by different procedures, a comparison of the above picric acid method was made with the following: In table 5, with a solution of the curd of butter in 50 per cent acetic acid; in table 6, with an aqueous suspension of the curd of butter concentrated on the steam bath; and in table 7, with a suspension of the curd in

TABLE 5

Showing amino nitrogen as per cent of total nitrogen in (A) picric acid and acetic filtrate; and (B) in curd of butter dissolved in 50 per cent acetic acid by heating for one hour at 50°C.

NUMBER	DESCRIPTION OF SAMPLE	AMINO NITROGEN AS PER CENT OF TOTAL NITROGEN		
		A	B	Difference
1	Sweet cream butter.....	1.3	10.4	9.1
2	Sweet cream butter.....	2.0	10.5	8.5
3	Sour cream butter.....	3.3	13.0	9.7
4	Sour cream butter.....	3.8	14.9	11.1
5	Neutralized sour cream butter.....	3.8	29.0	25.2
6	Neutralized sour cream butter.....	4.1	19.1	15.0
7	Neutralized sour cream butter.....	5.2	15.0	9.8
8	Neutralized sour cream butter no. 3, table 2.....	6.5	17.2	10.7
9	Sour cream butter.....	7.7	21.9	14.2
10	Old rancid butter.....	9.0	65.2	56.2

TABLE 6

Showing amino nitrogen as per cent of total nitrogen in butter: (A), picric acid and acetic acid filtrate; and (B), in suspension of curd washed from butter fat with hot water and concentrated on steam bath

NUMBER	DESCRIPTION OF SAMPLE	PER CENT OF TOTAL NITROGEN AS AMINO ACID		
		A	B	Difference
1	Sweet cream butter.....	1.6	11.5	9.9
2	Neutralized sour cream butter (no. 6, table 3).....	4.1	18.2	14.1
3	Sour cream butter no. 9, table 3.....	7.7	28.7	21.0
4	Neutralized sour cream butter.....	9.5	28.0	18.5
5	Rancid butter no. 10, table 3.....	9.0	45.4	36.4

TABLE 7

Showing amino nitrogen as per cent of total nitrogen of butter in: (A) picric acid and acetic acid filtrate; and (B) suspension of curd in water after distilling off ammonia under reduced pressure

SAMPLE NUMBER	DESCRIPTION OF SAMPLE	ALKALI USED IN DISTILLATION	PER CENT OF TOTAL NITROGEN AS AMINO NITROGEN		
			A	B	Difference
1	No. 3, table 3.....	Na_2CO_3	3.3	20.3	17.0
2	No. 3, table 2.....	MgO	6.5	33.0	26.5
3	Neutralized sour cream butter.....	MgCO_3	4.0	16.4	12.4

water after the ammonia had been removed by vacuum distillation. In every case the picric acid procedure gave lower results although there was a greater difference between samples of different quality by the picric acid method than by allowing nitrous acid to react on a suspension of the entire curd of the butter. There is a greater increase of amino nitrogen by the latter procedure on old butter than on fresh butter, which indicates that the protein of old butter is more readily hydrolyzed than that of fresh sweet cream butter. Therefore, when the amount of amino nitrogen present in a sample of cream or butter is used as an index of the quality of the sample, the determination in the acetic acid and picric acid filtrate will more nearly represent the amount actually present.

The advantages of the use of picric acid and acetic acid in preparing samples of milk, cream, buttermilk and butter for determination of the nitrogen reacting with nitrous acid in Van Slyke's apparatus as an index of proteolysis are:

1. Easy and rapid separation of the proteins and more complex substances from the lower degradation products, principally amino acids.
2. Hydrolysis of proteins during analysis is reduced to a minimum.
3. The filtrate can be held without further change, allowing a reasonable time between preparation of sample and analysis.
4. A correlation between amount of amino nitrogen and quality of sample.

By this procedure the amino nitrogen and ammonia in 14 samples of fresh butter from sweet cream was found to range from 0.9 to 2.3, with an average of 1.4 expressed as per cent of the total nitrogen.

THE DANGER OF IMPROPERLY CAPPED MILK BOTTLES¹

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INTRODUCTION

Except in the case of our certified milks, the protection that the dairyman and distributor can give the milk ceases when it is bottled and left by the driver on the consumer's doorstep; in fact, it can be said that the protection essentially ceases at the time when the milk is bottled. During the interval between the bottling and consumption of the milk there exists a period when the "safety bars" are let down, so to speak. I refer to the imperfect covering for the top of the milk bottle—the so-called slip-in type of bottle cap which leaves the lip of the bottle and the top entirely exposed. The writer has frequently noticed a bottle of milk on the family doorstep surrounded by flies and often a dog or cat licking the droplets of milk from the top. Aside from the repulsiveness of such occurrences, the rôle of flies in the transmission of typhoid and numerous other diseases is so well established that a discussion is unnecessary.

But probably most important, although least thought of, is the fact that the bottle is grasped around the top by the driver or other person handling it. The objection to this practice is obvious. Sanitarians have long insisted upon the danger of unclean hands of employees handling food stuffs, and in the case of a product so delicate and perishable as milk, this danger and objection is doubly serious.

The certified dairies have solved the uncovered bottle top problem by supplying a cover that slips over the outside, thus protecting the lip of the bottle over which the milk must pass

¹ Paper read at the annual meeting of the California Association of Dairy and Milk Inspectors, Santa Monica, California, September 28, 1921.

when being poured out. These covers are recognized as being ideal and very desirable, but as yet only a small portion of our milk is so delivered due to the considerable additional expense. The slip-in type of bottle cap is far more common due to the ease and simplicity of use and low cost. Probably 90 per cent of all bottled milk is served to the patron in this latter type of bottle and cap.

EXPERIMENTAL WORK

In order to ascertain the extent of contamination possible to the contents of the milk bottle when the lip of the bottle was artificially infected, the following work was carried out.

Protocol 1

Six half-pint bottles of milk, covered by inverting glass tumblers over the tops, were sterilized in the autoclave at twenty pounds steam pressure for thirty minutes.

The lip of each of three bottles was then infected by transferring a loop of a broth culture of streptococcus to the outer margin.

The infected spot on each bottle was wiped with a piece of sterile dampened gauze and 3 cc. of the milk poured from each bottle to a sterile test tube.

The three remaining uninfected bottles were each wiped with sterile dampened gauze and 3 cc. of the milk poured into sterile tubes (controls).

Results: After four days the test tubes of milk from infected bottles were solidly coagulated and showed the presence of streptococci. The test tubes of milk from the controls remained sterile.

Protocol 2

Same technique as outlined for protocol 1 except that *B. subtilis* (six days old sporulating) culture was substituted.

Results: Infected bottles all showed peptonization of the milk after seven days. Controls remained sterile.

The common custom of wiping the lip of the milk bottle with a damp dishcloth before taking out the cap suggested the use of the sterile gauze in protocols 1 and 2. The housewife generally believes that wiping with a dishcloth cleans the bottle. That the idea is a fallacy is shown by the fact that even the sterile gauze

failed to clean the neck from infection. The fact that the dishcloth will remove visible dirt no doubt accounts for the reliance generally given this much-used implement of the household. To prove that "visible-cleanness" does not necessarily run parallel with "bacteriological-cleanness" is demonstrated in the next experiment.

Protocol 3

Three sterile bottles were wiped with a dishcloth secured from an average household and impressions made of each by inverting sterile agar plates over the top of the bottles.

Results: All three plates gave heavy growths (fig. 1).

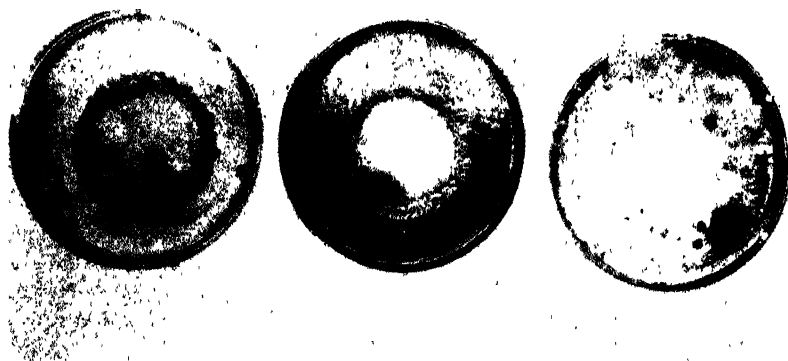


FIG. 1. BOTTLE TOPS WIPED WITH A DISHCLOTH; IMPRESSIONS MADE ON STERILE AGAR PLATES

A second set of exposures was necessary to get good pictures; consequently, some air-contaminating organisms are shown over the surface of the plates.

QUALITATIVE STUDY

It would be natural to assume that the bacterial flora adhering to the lip of an ordinary milk bottle might vary greatly. For the purpose of this work, however, identification of all species seemed unnecessary and consequently only the colon-typhoid group was investigated.

Protocol 4

Several bottles of milk were purchased on the market from different dairies and impressions made of each on petri dishes by resting an inverted plate of Endo agar on the top of the bottle.

Results: The average colon count for eight plates gave 21 (fig. 2).

No attempt was made in this examination to differentiate the members of the colon group.

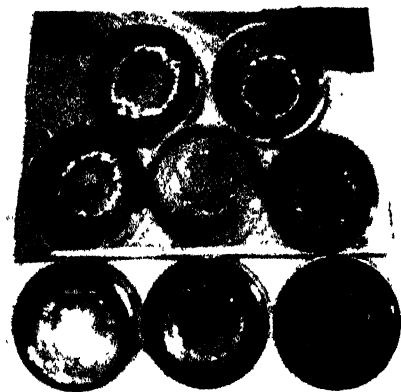


FIG. 2. ENDO AGAR COLON-AEROGENES GROUP

The small snapshots of the petri dishes did not bring out the growths satisfactorily, hence the prints have been retouched with white ink.

PROTECTIVE MEASURES

The work thus far shows that serious contamination from the lip of the bottle is a possibility. It occurred to the writer that some means of eliminating or controlling this condition would be valuable information. Aside from the use of the bottle-top-cover-cap (used chiefly by our certified dairies) which seems prohibitive for the general milk trade due to the expense, several methods came to mind:

Washing the neck of the bottle with hot water and soap would not be highly efficient at best and probably would be of little value, if done at all. Washing the lip of the bottle with a disin-

fectant could not be safely recommended for general use. "Burning off" the lip of the bottle over the free flame of a gas stove appeared practical and feasible.

Protocol 5

Three sterile bottles were infected with a broth culture of *B. subtilis*. Each was wiped with sterile gauze. The lip of bottle 1 was *not* subjected to "burning off." Bottle 2 was "burned off" by turning the bottle one complete revolution through the flame followed by rolling the lip of the bottle on the surface of a plate of solidified sterile agar. Bottle 3 was "burned off" by turning six times and rolling on the surface of an agar plate as above.

Results: Bottles 1 and 2 showed heavy growths; no. 3 showed none.

The results of the preceding experiment show that one complete turn of the bottle thru the flame did not materially reduce the infection of the lip of the bottle and that six complete turns did apparently render the lip sterile. In order to determine the least amount of burning necessary, impressions of the lips of a series of bottles of market milk were made on sterile agar plates; the lip of each bottle was then wiped with a damp dishcloth and another impression made on a sterile plate; finally the lip of each bottle was burned off by passing through the free flame of a gas stove while making one, two or three complete revolutions of the bottle and again making impressions on agar plates.

RESULTS

Most of the bottles gave less growths before being wiped, showing conclusively that the dishcloth either spreads the contamination or adds more to the bottle. Nevertheless, proper "burning off" even after wiping reduced the resultant growth to zero or a very negligible amount. Three turns of the bottles through the flame appeared sufficient in most cases.

CONCLUSIONS

1. Serious contamination from the lip of the milk bottle is possible.

2. A cap that covers the lip or rim of the bottle is undoubtedly the ideal protection.

3. In the absence of a cover-cap a practical protective measure is offered for use especially during outbreaks of communicable diseases.

RECOMMENDATIONS

1. "Burning off" the lip of the bottle by turning the bottle rapidly over the free flame of an ordinary gas cook stove will practically render the same sterile if done properly.

2. "Burning off" the lip of the bottle is practical, simple, inexpensive, and safe for any one to do.

3. The turning must be done quickly else the bottle may be cracked. The rule followed in this work has been to count 5 to 8 rapidly turning the bottle while counting. The lip only (not the entire neck) of bottle should be held in the flame.

4. By burning off the lip of the bottle as soon as the milk is brought into the house the cream will have ample opportunity to again set before the milk is to be used.

THE MILKING MACHINE AN IMPORTANT FACTOR IN MILK QUALITY

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For a good many years most of the fluid milk producers in New York State have had to produce milk in accordance with health board regulations which limit the number of bacteria in the milk at the time of delivery. Back of this is the assumption that milk, produced and handled so that its germ content is uniformly low, will be on the whole safer and better in quality than milk normally high in germ content. There is room for differences of opinion regarding certain exceptions to this statement, but, so far as the general milk supply is concerned, it unquestionably is true.

The outstanding requirement in practically all of these regulations is, that all milk must be classed into grades. In the grades, usually designated as A, B, or C, are specified more or less arbitrary standards governing the conditions under which the milk is to be produced and the number of bacteria allowed for each grade. The latter requirement is the important one because of its true relation to actual quality, and constitutes one of the big problems confronting the producers of fluid milk, particularly. The force of this statement is appreciated when one considers the bacterial requirements for each grade as established, for instance by the New York City Board of Health. Milk, to qualify as grade A, raw, "shall not contain more than thirty thousand (30,000) bacteria (colonies) per cubic centimeter when delivered to the consumer or at any time prior to such delivery." Milk to qualify for pasteurization as grade A in the country but to be sold in the city "shall not contain more than one hundred thousand (100,000) bacteria (colonies) per cubic centimeter at any time before pasteurization." The limit for milk to be pasteurized as grade B is 300,000 per cubic centimeter. In order to insure the low counts specified in grade A, many

commercial fluid milk dealers offer to the patrons a certain premium per hundred weight for milk delivered to their plant with a count under 10,000 per cubic centimeter (agar plate counts). A smaller premium is paid if the count is between 10,000 and 25,000 per cubic centimeter. These premiums are sufficiently great to be well worth striving for; consequently, the influence of any dairy operation upon the germ content is of economic importance to the dairymen.

No small amount of difficulty is encountered by the dairymen in meeting these bacteriological requirements. During March, 1921, the Dairy Department of the New York State College of Agriculture instituted an educational program of extension work designed to bring directly to the producers of milk, information concerning the sources of bacteria in milk and their relationship to quality. The method of procedure will not be given here since it has already been discussed in detail in the JOURNAL. The work is based upon a bacteriological study, by means of the direct microscopic method of the milk as delivered by the patrons at any given plant on two or three successive days. The results are presented and discussed at a previously advertised meeting of the patrons. The grading of the milk in this way makes it possible to collect valuable information regarding the general quality of the milk as delivered by the dairymen and the most important factors affecting the bacteria content. From March until September, inclusive, the quality of milk at fifteen fluid milk plants has been studied in this way.

High bacteria counts in milk are due to excessive contamination, primarily from the things with which the milk comes in contact, or to improper cooling, or to both combined. In studying milk under the microscope it is possible to note the general types of bacteria present and to roughly classify them according to their probable source. For instance, a predominance of lactic-acid-producing types in the milk as delivered by a patron, indicates lack of cooling. A predominance of organisms which form large clumps indicates utensil contamination. Long-chained streptococci associated with leucocytes serve as a basis for suspecting some form of infections target. Recent observa-

tions on milk machine contamination have revealed the fact that the predominant flora of machine-drawn milk frequently consists of a mixture of the large clump-forming micrococci and the long-chained streptococci, which are, at least morphologically, identical with the forms commonly associated with garget. By tracing back individual cases to the farm, it has been proved beyond doubt that improperly cleaned milking machines may harbor the long-chained forms in sufficient numbers to yield an initial contamination to a full can of milk of over 1,000,000 groups per cubic centimeter. It would not do to assume that these long-chained forms are the causative agents of infectious garget. The resemblance may be purely morphological, but their presence serves as a guide to the source of contamination, and until more is known as to their exact identity they should be regarded as a possible source of an outbreak of garget in the herd. In several instances a thorough cleaning of the machine eliminated them, together with micrococci forms, to the extent that none could be found after searching as many as one hundred fields of the microscope. This classification of types as regards their possible origin is not absolute by any means, but is sufficiently so, to be of great assistance in locating the cause of high counts.

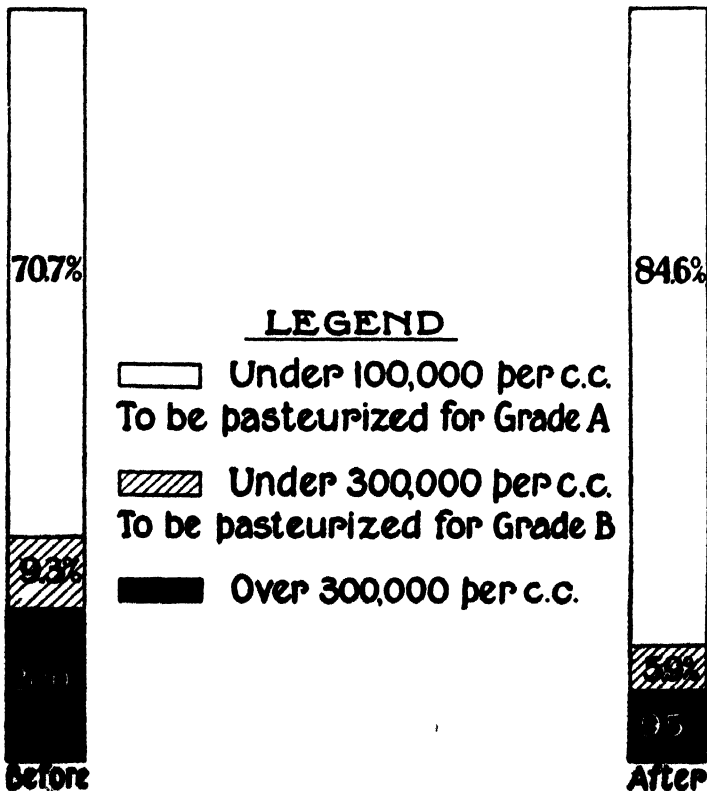
Thus far the observations made in grading 3243 samples from milk delivered by 1104 patrons have led to the conclusion that the commonest causes of high bacteria counts in milk at the time of delivery to the plant are: (1) failure to promptly and properly cool; (2) high contamination of milk by cans and other utensils which have not been thoroughly steamed or scalded, or promptly dried, and (3) high contaminations from dirty milking machines. There is little question that if these three factors were properly controlled the amount of milk high in bacteria count, due to other possible sources of contamination, would be insignificant in comparison to the total quantity delivered.

The data collected in these studies have made it possible to accumulate fairly accurate information concerning the bacterial quality of machine-drawn milk as compared to hand-drawn milk. The observations on the bacterial quality of milk as affected by milking machines were made at twelve of the fifteen plants,

comprising 790 patrons, of which 635 were hand milkers and 155 were users of milking machines.

Wherever possible, it is highly important that the milk at each plant be again graded after the meeting of the patrons as well as

AVERAGE QUALITY OF 2001 SAMPLES OF HAND DRAWN MILK BEFORE ^{AND} AFTER MEETING OF PATRONS



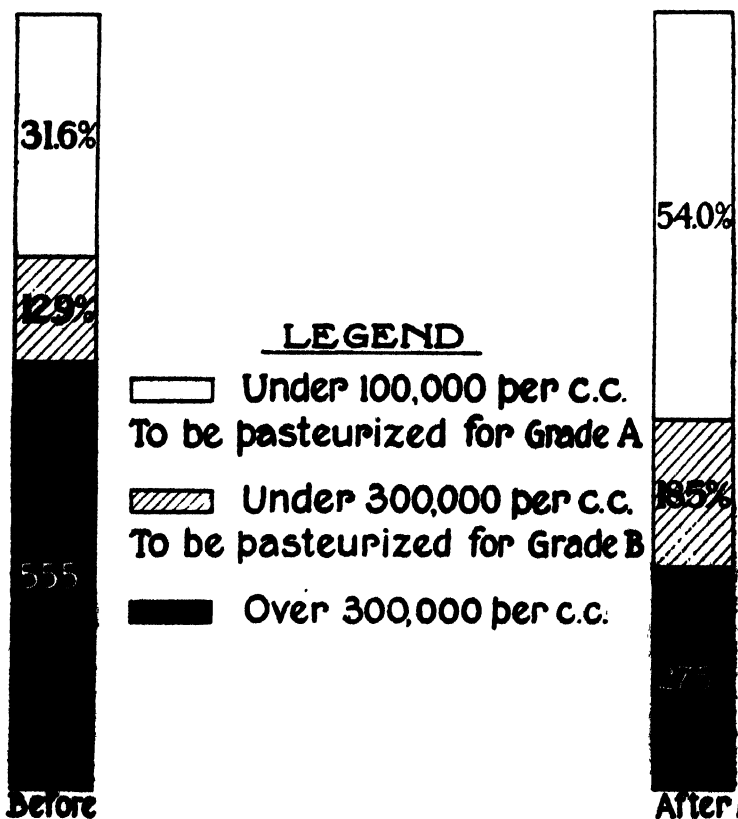
GRAPH 1

before, in order to determine the extent of improvement made as a result of putting into effect some of the recommendations made. In graph 1 is shown the percentage rating of 2001 samples of hand-drawn milk before and after the meetings of the patrons.

In graph 2 is shown the percentage rating of 548 samples of machine-drawn milk before and after the meetings of the patrons.

The picture presented in these graphs clearly indicates that the milking machine contributes a large share toward high bac-

AVERAGE QUALITY OF 548 SAMPLES OF MACHINE DRAWN MILK BEFORE ^{AND} AFTER MEETING OF PATRONS



GRAPH 2

teria counts. There is good reason to believe that the cooling and other dairy operations are done equally well by both classes of milkers; but the difference between the amounts of milk

delivered by each that contained more than 300,000 bacteria per cubic centimeter is so great as to leave no doubt that the milking machine, as it is cleaned and cared for on the average farm, is a serious source of contamination. In fact, approximately three times as much inferior milk was delivered by users of machines as by hand milkers.

Considerable opposition has developed in dairy sanitary circles against the use of milk machines because so much milk inferior in quality has been delivered by the users. In too many instances this opposition has assumed the proportions of a prejudice against their use in general. This is particularly unfortunate, because due to labor conditions, the milking machine has undoubtedly become an essential part of modern dairy equipment. Instead of its decreasing in general use, as some think will be the case, there is every reason to believe that there will be an increase.

There is little question that the modern milking machine, from a mechanical point of view, will satisfactorily draw the milk from the cow if it is operated according to directions. Also the statement cannot be questioned that the milking machine, if properly operated, eliminates to a very large degree much of the unpleasantness so characteristic of hand milking. Furthermore, because of the fact that one man can normally milk from twenty to twenty-five cows per hour, requiring less average time per cow than is required in hand milking, the milking machine is an important economic factor in the production of milk, especially with large herds.

At present the chief limiting factor in the successful operation of milking machines is the lack of recognition on the part of many dairymen and machine agents of the importance of proper cleaning. If measures are not taken fairly promptly, by all concerned, to make the general quality of machine-drawn milk equal to that drawn by hand, there is danger of a reaction against a very useful and practical method for milking cows.

Not infrequently the question is raised as to whether it is possible to produce a high-grade milk where machines are used. Abundant evidence exists to answer this affirmatively. Producers of certified milk are successful in meeting the bacterial

requirements, as are also many dairymen who sell milk where premiums are paid if the bacteria count averages under 10,000 per cubic centimeter. At the Clover Farms Dairy Company's Plant at Homer, New York, where milk has been bought on this basis for a number of years, 54 per cent of the twenty-six users of machines during April, May and June, 1921, produced milk with a count under the 10,000 per cubic centimeter according to the counts made by the Company, and 23.7 per cent produced milk with a count between 10,000 and 25,000 per cubic centimeter. Only 6.6 per cent produced milk containing over 300,000 bacteria per cubic centimeter. The hand milkers, however, did somewhat better, 74.7 per cent being under the 10,000, 12.1 per cent between 10,000 and 25,000 and only 3.5 per cent producing milk with more than 300,000 bacteria per cubic centimeter. While the advantage appears to be in favor of the hand milkers, yet in the case of the users of milking machines the 54 per cent constitutes sufficient evidence to indicate what can be done if strict attention is given to all details. As shown by the results pictured in graph 2, substantiated by observations in the field, the production of milk at least good enough in quality to qualify for pasteurization as grade A is a comparatively simple matter if attention is paid to a few essentials. Referring to graph 2, the 55.5 per cent over 300,000 before the meeting of the patrons and the 27.5 per cent after the meeting are the result of gross carelessness or misinformation. Much of this, of course, may be traced to the dairyman's lack of information regarding bacteriological relationships, and until some appreciation of the importance of the common sources of bacterial contamination is acquired, the users of milking machines will continue to lag considerably behind the hand milkers so far as bacteria counts of milk are concerned.

The milking machine, with its more or less complicated mechanism—pulsators, heavy pail and lid, valves, rubber rings, moisture traps, rubber tubing, inflations, test cups; and so on, with most of which the milk comes directly in contact—constitutes a fertile source of contamination unless special precautions are observed. Among the first considerations before installing any machine

THE CAUSES OF LEAKY BUTTER

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INTRODUCTION

The moisture and salt content of most butter packed and stored in the usual way gradually decreases as the butter becomes older. The brine comes to the outside of the butter and either runs away or evaporates. This movement of brine from within the butter to the outside is commonly spoken of as leakage, and butter in which this movement is especially pronounced is called leaky butter. Leaky butter loses rapidly in weight due to the loss of brine, and this loss in weight is known as shrinkage. When leaky butter is cut into prints considerable brine is forced out which results in a marked decrease in weight.

Wet looking butter is butter that shows much free brine on its surface in the churn, and especially when a sample is taken with a butter trier after the butter has hardened in the refrigerator. Wet looking butter is commonly called leaky although no experimental work has been done to show that the two are the same.

A review of the dairy literature reveals the fact that no systematic attempt has ever been made to determine all of the conditions of manufacture that make butter leaky. Text books on butter making give a variety of reasons for the causes of leaky butter. McKay and Larsen, in their book, *Principles and Practices of Buttermaking*, state: “. . . this leaky condition is brought about chiefly by churning the butter to small granules, washing the butter very little in cold water, salting heavily while butter granules are still small and firm, and working the butter frequently in the presence of brine.” Martin H. Meyer in his

¹ This work was done by the author under the supervision of Prof. E. H. Farrington at the University of Wisconsin.

Modern Buttermaking discusses "leaky or slushy butter" as being caused by not cooling the cream sufficiently before churning, or by churning it too soon after it is cooled. It may also be caused by washing the butter with too warm or too cold water. Slushy butter is more apt to appear in the spring than at any other time of the year. The Book of Butter by E. S. Guthrie, states, "Often the butter is not worked sufficiently to incorporate the moisture properly, in which case the water collects in pockets. When this butter is cut, it appears leaky." O. F. Hunziker, in his book The Butter Industry discusses leakage of butter more extensively than the other writers. The conditions which he gives as the causes of leaky butter are too high a churning temperature, churning too quickly after cooling to churning temperature, insufficient working, rich cream, and tearing the butter apart rather than squeezing it together during working. This author also states: "Within relatively narrow limitations leakiness may be intensified or minimized by the processes of washing, salting, and working, but the fundamental cause of leakiness lies prior to these processes; it has to do with the treatment the cream receives preparatory to the churning process."

METHOD USED TO DETERMINE LEAKINESS OF BUTTER

The butter was packed out of the churn in so-called leakage tubes. The leakage tube consisted of a cylindrical tin can holding 28 pounds of butter which was wide open at one end and tapered like a funnel at the other. A 50-cc. burette was attached to the funnel end by means of a rubber stopper, and the lower end of the burette was plugged with a rubber stopper. The brine was caught in the burette and was measured as rapidly as it ran out of the butter. The upper part of the butter was cut level with the top of the tin can and was sealed by means of a large sheet of wrapping paper dipped in hot paraffin, which was placed on the butter before the paraffine hardened. The leakage tubes were, therefore, water-tight and the brine drained off the butter as rapidly as it leaked out. Readings in cubic centimeters of brine were taken for a period of seven days. Experiments proved that this was sufficient time in which to obtain repre-

sentative results. By the use of these leakage tubes a leakage of 1 cc. of brine or approximately 0.0025 of a pound could be determined, and this method was far more sensitive than the old method of weighing the butter to the tenth of a pound.

The effect of each variation in the manufacture of butter on its leakiness was compared with a normal or check churning from the same batch of cream churned in the same churn, a Perfection No. 6. Half of the cream in the vat was churned in the usual way, and two leakage tubes were packed. The remaining half was churned with the variation in handling the cream or butter which was being tried for its effect on leakiness. Two more leakage tubes were packed from this butter. The results of three sets of churnings, six churnings in all, from three different vats of cream and all determining the effect of one factor were considered sufficient evidence on which to draw conclusions.

EFFECT OF SIZE OF CHURNING ON LEAKINESS OF BUTTER

The cream in this experiment was divided into two unequal sized portions which were churned in the same identical manner. The larger churnings were normal sized and the smaller ones contained just enough butter to completely cover the shelf in the churn and be properly worked. Table 1, giving the weight of butter in each churning with the leakage expressed as cubic centimeters of brine and also as the shrinkage per hundredweight of butter, is a brief summary of the results obtained.

It is evident from table 1 that the butter from the small sized churnings was uniformly less leaky than butter from the larger churnings. The less leaky butter also appeared drier in the churn. The butter from vat no. 30 leaked twice as much as the butter from vat no. 28. No attempt was made to hold all conditions uniform from day to day, and a comparison of the butter made from the various vats of cream with each other is not justifiable. The butter from one churning can be compared only with its counterpart from the same vat of cream.

TABLE 1

Effect of size of churning on the leakiness of butter

VATS OF CREAM	WEIGHT OF BUTTER	SEVEN DAYS LEAKAGE	
		Brine	Shrinkage per hundred weight of butter
	<i>pounds</i>	<i>cc.</i>	<i>pounds</i>
No. 28.....	270	71.4	0.604
	460	106.5	0.900
No. 29.....	270	94.1	0.799
	498	140.8	1.193
No. 30.....	211	131.3	1.110
	348	187.8	1.586
Average.....	250	99.9	0.846
	435	145.0	1.228

TABLE 2

Effect of churning temperature on leakiness of butter

VATS OF CREAM	CHURNING TEMPERATURE	SEVEN DAYS LEAKAGE	
		Brine	Shrinkage per hundred weight of butter
	<i>°F.</i>	<i>cc.</i>	<i>pounds</i>
No. 12.....	55	32.5	0.275
	50	32.2	0.271
No. 13.....	54	4.5	0.038
	48	42.9	0.368
No. 14.....	54	25.0	0.211
	48	7.0	0.059
Average.....	54	20.7	0.175
	48	27.4	0.231

EFFECT OF CHURNING TEMPERATURE ON LEAKINESS OF BUTTER

One-half the cream was churned at the usual temperature and the other half was churned 6° too warm. The butter was washed at such a temperature that the finished butter would be of the

same temperature and hardness regardless of the temperature of the cream when it went into the churn. The results obtained are given in table 2. They show that although the butter churned warm did not look very much more wet in the churn than the butter from the colder churnings, the churning temperature had no effect on the rate of leakage. The churning temperature is a negligible factor in causing leaky butter provided the butter is washed at such a temperature that the butter has the proper firmness during working.

EFFECT OF TIME HELD COLD AFTER PASTEURIZATION ON LEAKINESS OF BUTTER

After pasteurizing the vat of cream one-half was churned as soon as the cream was at the churning temperature and the other half was held cold over night, or about twenty hours, before it was churned. The butter from the cream churned without having been held cold was very soft and warm so it was necessary to wash it much colder in order that the temperature of the finished butter would be the same as that of the butter churned from the cream which was held cold for twenty hours. The results set forth in table 3 show that although the butter made from the cream churned without holding cold after pasteurization looked very much more wet in the churn, the actual leakage was not any greater than that of the butter held cold for twenty hours after pasteurization.

EFFECT OF TEMPERATURE OF WASH WATER ON LEAKINESS OF BUTTER

The two churnings from the same vat of cream were handled in the same manner excepting that the wash water of one was 10°F. warmer than that of the other. This resulted in 5° difference in the temperature of the finished butter which was not enough to injure the body of the warmer butter. The data giving the results of this experiment are summarized in table 4. The butter washed warm was very soft when made; it appeared a little more wet in the churn, but held its moisture on standing

much better than the butter washed cold. This result is directly opposed to present opinion, but these results and others obtained on the shrinkage of butter in cold storage prove conclusively

TABLE 3

Effect of time held cold after pasteurisation on leakiness of butter

VATS OF CREAM	HELD COLD	SEVEN DAYS LEAKAGE	
		Brine	Shrinkage per hundred weight of butter
	<i>hours</i>	<i>cc.</i>	<i>pounds</i>
No. 31.....	0	141.1	1.193
	20	128.6	1.088
No. 32.....	0	105.0	0.888
	20	100.5	0.850
No. 33.....	0	8.9	0.075
	20	3.9	0.033
Average.....	0	84.7	0.716
	20	77.7	0.657

TABLE 4

Effect of temperature of wash water on leakiness of butter

VATS OF CREAM	TEMPERATURE OF WASH WATER	SEVEN DAYS LEAKAGE	
		Brine	Shrinkage per hundred weight of butter
	<i>°F.</i>	<i>cc.</i>	<i>pounds</i>
No. 9.....	54	137.2	1.161
	65	28.9	0.245
No. 10.....	55	165.8	1.403
	65	34.8	0.298
No. 11.....	55	119.2	1.008
	65	26.5	0.224
Average.....	55	144.9	1.226
	65	30.0	0.254

that butter which is soft when worked holds its moisture much better than hard butter, providing both are worked the same number of revolutions.

In the experiments to determine the effect of the time the cream was held cold after pasteurization before churning, and the effect of the churning temperature on leakiness, the temperature of the wash water was such that the finished butter always was the same hardness. If the wash water had been used at the same temperature regardless of the condition of the butter, then a warm churning temperature and churning immediately after pasteurization would have resulted in a soft butter that would have been less leaky. If no variation is made in the amount the butter is worked the common opinion that churning immediately after pasteurization, churning at a warm temperature, and washing butter warm causes butter to be leaky because it is wet looking is incorrect.

EFFECT OF THE AMOUNT OF WATER IN THE CHURN DURING WORKING OF THE BUTTER ON ITS LEAKINESS

The leakiness of butter worked in a dry churn was compared with butter worked in brine. The churn was made dry by placing a short piece of cork in each door so that the doors when closed remained about $\frac{1}{4}$ inch open. Whenever the churn contained water it was stopped with the doors on the under side and all water was drained out. To the butter worked in water $2\frac{1}{2}$ pails of strong salt brine of the same temperature as the butter were poured into the churn and the doors were tightly closed. The butter worked in water contained more moisture, looked more wet in the churn, and the results stated in table 5 show that it was more leaky than butter worked in a dry churn. This confirms the practice of many creameries of finishing the working of the butter in a dry churn for the purpose of making the butter non-leaky.

EFFECT OF AMOUNT BUTTER IS WORKED ON ITS LEAKINESS

When the butter had been worked just enough to remove all grittiness and prevent mottles, two leakage tubes were filled. The remaining butter was worked until it appeared close grained and waxy, and two more leakage tubes were filled. This experi-

ment should not be interpreted as indicating the correct number of revolutions that butter should be worked, for commercial practice has demonstrated that the amount of working butter

TABLE 5

Effect of amount of water in the churn during working of the butter on its leakiness

VATS OF CREAM	BRINE IN CHURN	SEVEN DAYS LEAKAGE	
		Brine	Leakage per hundred weight of butter
	<i>gallons</i>	<i>cc.</i>	<i>pounds</i>
No. 23.....{	0	3.8	0.032
	7	104.0	0.980
No. 24.....{	0	40.4	0.342
	7	53.3	0.451
No. 25.....{	0	87.7	0.666
	7	175.5	1.485
Average.....{	0	40.9	0.346
	7	110.9	0.938

TABLE 6

Effect of amount butter is worked on its leakiness

VATS OF CREAM	REVOLUTIONS WORKED	SEVEN DAYS LEAKAGE	
		Brine	Leakage per hundred weight of butter
		<i>cc.</i>	<i>pounds</i>
No. 6.....{	20	98.6	0.834
	40	0.1	0.001
No. 7.....{	20	61.2	0.518
	30	0.7	0.006
No. 8.....{	20	180.1	1.524
	30	1.1	0.009
Average.....{	20	113.3	0.958
	33	0.6	0.005

should receive is variable. The amount this experimental butter was worked represents the minimum rather than the maximum working that butter should receive.

The data presented in table 6 give very contrasting leakages. The butter worked the greater number of revolutions looked very dry in the churn and held its moisture exceedingly well. As a result of observations on all the churnings reported in this paper it became very evident to the author that the amount butter is worked has a greater effect of leakiness than any other individual factor, and butter which might otherwise be leaky could be made non-leaky by working it as much as possible without injury to the body of the butter.

All of the butter made in this experiment was scored by competent butter judges and no criticisms were made of the body, color or salt. One must conclude that either (1) the butter judges were very lenient in considering the body of the butter because the butter which was made from vat no. 8 and worked 20 revolutions lost 1.5 pounds in seven days, while the butter from the same vat of cream that was worked 30 revolutions lost only 0.009 pounds in seven days, or (2) the butter judges could not distinguish between leaky and non-leaky butter.

EFFECT OF STORAGE TEMPERATURE ON LEAKINESS OF BUTTER

Four leakage tubes were filled from the same churning, two of which were held in a warm room and two in the refrigerator. The data contained in table 7 gives conclusive evidence that butter held at a cold temperature leaks brine more rapidly than if held at a warm temperature. This fact was very noticeable throughout the entire work, and it is one of the reasons for an uneven leakage from day to day. Whenever the refrigerator became very cold the leakage was always greater, and the leakage could be almost stopped by a warm storage temperature. It must be remembered, however, that the temperatures employed were not cold storage temperatures, and the results obtained do not necessarily apply to freezing temperatures. If this experiment had been so conducted that evaporation took place from the butter it is very probable that different results might have been obtained.

COMPARISON OF LEAKINESS OF BUTTER PRODUCED IN SUMMER
AND WINTER

Churnings of butter produced under winter conditions were made to compare with churnings of butter produced under summer conditions. The winter butter was produced in the early part of April and the cattle were fed hay, grain, and silage. The summer butter was produced in June and the cattle received little or no supplement to the pasture. In every churning the finished was the same number of degrees above the churning temperature and as firm in the summer as in the winter.

TABLE 7
Effect of storage temperature on leakiness of butter

VATS OF CREAM	TEMPERATURE OF STORAGE	SEVEN DAYS LEAKAGE	
		Brine	Leakage per hundred weight of butter
	°F.	cc.	pounds
No. 20.....	45	12 7	0 107
	63	0 6	0 005
No. 21.....	48	35 0	0 296
	64	4 1	0 035
No. 22.....	48	89 0	0 753
	65	3 0	0 025
Average.....	47	45 6	0 387
	64	2 6	0.022

The results of these comparisons gave a slightly greater total leakage for the summer butter. It was difficult to duplicate conditions exactly with a spread of two months between churnings and individual variations between churnings were so great that the differences obtained in the total leakage became insignificant. It must be concluded therefore that little or no difference exists between the leakage of summer and winter butter when only the composition of the butter is concerned. The results of other experiments reported in this paper indicate that the heavy shrinkage in butter often noted in commercial work



FIG. 1. LEAKY AND NON-LEAKY BUTTER PACKED INTO ASH TUBS

Note the water logged and stained condition of the tubs at the left containing leaky butter as contrasted with the drier, whiter tubs at the right containing non-leaky butter. This difference in the appearance of the tubs is usually no index of the leakiness of the butter they contain because tubs vary so greatly in their absorptive powers.

during the spring months is probably caused by the use of cold wash water and a small amount of working usually employed to prevent a salvy, greasy body and to lower the moisture content of the finished butter.

EFFECT OF SALT CONTENT OF BUTTER ON ITS LEAKINESS

An excessive amount of salt was added to the first churning and an average amount was added to the second churning from the same batch of cream. None of the butter was gritty. The data summarized in table 8 show that the highly salted butter leaked brine about twice as rapidly as the butter which contained a smaller amount of salt.

TABLE 8
Effect of salt content of butter on its leakiness

VATS OF CREAM	SALT	SEVEN DAYS LEAKAGE	
		Brine	Leakage per hundred weight of butter
		cc.	pounds
No. 15.....	3 8	283 0	2 394
	2 2	166 0	1.404
No. 16.....	5 0	325 0	2.749
	2 7	170 0	1 442
No. 19.....	3 7	228 3	1.931
	2 1	46 6	0.392
Average.....	4 1	269 5	2.280
	2 3	127.5	1.079

EFFECT OF MOISTURE CONTENT OF BUTTER ON ITS LEAKINESS

It was practically impossible to make two churnings from the same vat of cream in such a way that there would be a marked difference in the moisture content of the finished butter without introducing some variation in the method of manufacture which would also affect the leakiness of the butter. Consequently, to determine the effect of the moisture content of butter on its leakiness all the churnings previously made were divided into

two groups. The leaky butter group includes those which were the more leaky of the two churnings from the same cream. The non-leaky butter, was that which leaked the least. All butter which showed no difference in the leakage from duplicate churnings were not included in the data. Table 9 summarizing the data is based on an average of thirty churnings.

The moisture content of the butter did not vary according to the leakiness. One sample containing 13.2 per cent of water was leaky, another containing 15.8 per cent was dry, and the average moisture content of the two lots of butter was quite uniform.

TABLE 9

Relation of moisture content of butter to its leakiness

	AVERAGE MOISTURE CONTENT	HIGHEST MOISTURE CONTENT	LOWEST MOISTURE CONTENT
Leaky butter.....	14.8	16.2	13.2
Non-leaky butter.....	14.3	15.8	13.3

IS WET LOOKING BUTTER LEAKY?

That butter which looks wet does not necessarily contain a high percentage of moisture has been shown in the past and the work was again repeated. Butter entered in a Wisconsin scoring exhibition was classified according to the free moisture noticed when samples were taken with a trier and it was then tested for moisture and salt. Table 10 summarizes the results.

No relationship exists between the percentage of water in butter and its appearance in respect to visible moisture. However, dairymen have always assumed that butter which looks very wet is also very leaky. The two terms are used interchangeably. The author previously thought this belief was correct and unfortunately did not in the beginning of this experimental work attempt to determine if butter which was leaky actually showed a wet trier sample. The methods of manufacture which causes butter to look wet have been observed in practice and table 11 compares these causes with the causes of leaky butter found as a result of this experimental work.

Factors which cause butter to look wet do not necessarily cause it to be leaky. Therefore wet looking butter is not always leaky, although they are doubtless closely associated with each other.

Butter looks wet because some of the moisture in it exists in large droplets and in pockets or holes in the butter. On a

TABLE 10
Relation of moisture content of butter to its apparent wetness

HOW THE BUTTER LOOKED	NUMBER OF TUBS	WATER	SALT
		<i>per cent</i>	<i>per cent</i>
Very dry.....	13	14.4	2.0
Dry.....	56	14.1	2.1
Little wet.....	51	14.2	2.1
Wet.....	12	14.6	2.2
Very wet.....	9	13.2	2.2
Average.....	141	14.1	2.1

TABLE 11
Comparison of the causes of wet looking and leaky butter

FACTOR TRIED	EFFECT ON LEAKAGE	EFFECT ON APPEARANCE
Large churning.....	Little leaky	No effect
Warm churning temperature.....	No effect	Wet
Not held cold after pasteurization.....	No effect	Wet
Warm wash water.....	Dry	Wet
Working butter in water.....	Leaky	Wet
Under working butter.....	Leaky	Wet
Warm refrigerator.....	Dry	Wet
High salt content.....	Leaky	Little wet
Butter high in soft fat.....	No effect	Wet
High moisture content.....	No effect	No effect

freshly cut surface this water can readily be seen and it is forced out by a trier when taking a sample. Butter is leaky because it has an open texture, or, in other words, is full of small capillaries through which the water can pass. The moisture which leaks out of butter does not simply run out like water running in a stream, but the moisture moves by capillarity. This is shown by the fact that after heavy leakage the butter always contained a

uniform moisture content throughout the entire mass, the percentage of water being the same at the top, bottom, sides and middle of the butter. Conditions in manufacture which made the butter-fat globules hard and preserve them in their natural spherical shape tends to keep the spaces between the globules as large as possible. Washing butter cold and underworking give such results. If butter is stored in a cold refrigerator the butter fat globules contract and the spaces between them must become larger. Hence, the butter becomes more leaky. Although some factors, such as a high salt content, which cannot affect the size of the capillarities did materially increase leakage, their effect can be almost entirely eliminated by reducing the size of the capillarities to such an extent that all leakage practically ceases. Their effect must, therefore, be considered of secondary importance.

PRACTICAL APPLICATION OF RESULTS OBTAINED

During the summer months butter was carefully made for cold storage with variations in the method so that the leakage would be affected. The same vat of cream was often divided and churned to make one-half of the butter leaky and the other half non-leaky. The butter was packed into paraffined 63-pound ash tubs. It was weighed net to the tenth of a pound as it came from the churn, and was re-weighed in like manner after four to seven months in cold storage. Table 12 summarizes the results. The results obtained with cold storage butter agree with those obtained by the leakage tubes. It is especially noteworthy that as small a difference as ten revolutions in the amount the butter was worked caused a variation of 100 per cent in the rate of shrinkage or leakage. The butter washed with very cold water leaked about five times as much brine as the butter washed with very warm water and the rate of leakage of butter washed at an average temperature was between these two extremes. The moisture content of some of the leaky butter decreased as much as 4 per cent while most of the drier butter lost about 1 per cent moisture.

It has been shown that various conditions affect the leakiness of butter. Some conditions which make butter retain its moisture tend to produce butter of poor quality and it is not advisable to injure the butter in any way to make it less leaky. Other conditions which make butter non-leaky cannot be practically used. The working of butter influences the leakiness to the greatest extent and should be used as the means of controlling shrinkage. Wash butter with a cold water so that the body will be medium firm, even though this caused it to be more leaky. Such butter needs much extra working to give it the proper waxy body, and when it has been worked sufficiently it will be of good body, close grained, dry looking and will retain its moisture

TABLE 12

Variations on shrinkage of cold storage butter

VARIATION IN MANUFACTURING	NET WEIGHT BEFORE STORAGE	NET WEIGHT AFTER STORAGE	LOSS	
			pounds	per cent
Average of 234 tubs.....	14,655.4	14,435.5	219.9	1.50
Normal temperature of wash water.....	9,577.4	9,447.8	129.6	1.35
Very cold wash water.....	831.4	795.9	35.5	4.27
Very warm wash water.....	1,425.1	1,414.5	11.2	0.78
Worked normal number of revolutions..	1,440.5	1,411.5	29.0	2.01
Worked ten more than normal.	1,380.4	1,365.8	14.6	1.05

better than if made in any other way. The extra working causes it to hold its moisture and is permitted without injury to the body of the butter, by the use of cold temperatures.

In judging the leakiness of butter by a trier sample, two conditions should be noted: first, the openness of texture, and second, the amount of free brine. Close grained butter is not leaky even though it may appear wet. An open texture and much free brine indicate a leaky butter, but the amount of free brine alone is not a satisfactory index of the leakiness of butter.

CONCLUSIONS

Some of the results obtained from the experimental work reported in this article are contradictory to text book teachings

and the opinions of many practical creamery men. For that reason the conclusions drawn from the data presented are enumerated and briefly summarized as follows:

1. The leakiness of butter is not affected by (1) the churning temperature, (2) time held cold after pasteurization previous to churning, (3) the moisture content of the finished butter, (4) high percentage of the soft butter fats.

2. Butter is made leaky by (1) large churnings, (2) cold wash water, (3) working butter in water, (4) not enough working after salting, (5) high salt content, (6) cold refrigerator.

3. The amount butter is worked has a greater influence on leakiness than any other factor and should be used to control leakiness.

4. Wet looking butter is not always leaky.

5. The openness of texture is the best indicator of the leakiness of butter.

6. Butter made leaky lost 4.2 per cent in weight during a period of six months in cold storage and butter made non-leaky lost only 1 per cent in the same length of time.

THE INFLUENCE OF CERTAIN FACTORS ON THE METHYLENE BLUE REDUCTION TEST FOR DETERMINING THE NUMBER OF BACTERIA IN MILK¹

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One of the factors, and possibly the most important in determining the quality of milk is its bacterial content, hence, wherever a classification of milk is desired, a bacterial examination is essential. In the dairy manufacturing field, the payment for milk according to its bacterial content has made little headway. The future must bring this change into the industry if the quality of the products is to be maintained at even its present low level, to say nothing of the improvement that should take place. There is no reason why the same price should be paid for high quality milk, as for a milk that is sure to lower the grade of the product for which it is to be used. In the butter industry and in the market milk business certain curative measures can be employed to overcome the negligence of the producer in the production and handling of milk and cream. In the cheese industry, however, prevention, not cure, must be used. A difference in remuneration to the producer is the most powerful lever that can be employed to raise the quality of the milk no matter for what purpose it is intended.

Milk low in bacteria is, generally speaking, of higher quality for cheese making than is milk containing many bacteria. If the cheesemaker seeks to differentiate the milk of his patrons into grades, he must have some means of measuring the bacterial content. Again in the control of market milk the smaller cities have not kept up with the larger in the improvement of their

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milk supplies. They have not been financially able to duplicate the methods of the larger cities. No simple and inexpensive method of determining the bacterial content of milk has been available until recently, and for this reason progress under many conditions has been difficult.

The methylene blue reduction test is the only one that seems to meet the necessary requirements, in so far as simplicity and expense are concerned, for use in many fields, especially in the field of dairy manufacturing. This test is based on the fact that the color imparted to milk by the addition of the dye, methylene blue, disappears more or less quickly, the time required for its disappearance depending on a number of factors, most important of which are the bacterial content of the milk and the temperature at which the test is made. If all controlling factors can be kept constant other than the bacterial content of the milk, the time required for the color to disappear will be determined by the number of bacteria.

The great service which such an inexpensive and simple test might yield to the dairy industries of this country has led the authors to review the literature and to study the test from many points of view. This paper presents simply a summary of our work. In connection with nearly every point considered, many tests have been made. Our conclusions are, therefore, based on much more numerous data than it is possible to present here. It is hoped that this paper may serve to bring the reduction test to the attention of many who may find use for it.

In spite of the widespread use of the reduction test in Scandinavian countries and in other parts of Europe, the method has not been adopted by the dairy industry of this country. Apparently the chief reason for its non-adoption is the belief that it is at best a rough method. The main basis for this conclusion is the fact that the results obtained with it do not correlate exactly with those supplied by the plate culture method. Rahn (1920) has recently emphasized this point and has concluded that the method is not an accurate one. Some of the reasons for the non-correlation of the two methods were given by Hastings (1919). Others will be presented in this paper.

EXPERIMENTAL WORK

The effect of the concentration of methylene blue on the reduction time

In the practical application of the methylene blue reduction test varying concentrations of the dye have been used. Fred (1911) used 1 part of the crystalline dye to 20,000 parts of milk, Jone (1915) 1 part to 50,000; Mueller (1906) 1 part to 40,000; and Kufferath (1919) 1 part to 3000-4000. The latter noted that sterilized milks, by which expression was meant milks heated to the boiling point but not actually sterile, did not reduce the dye. He also noted that raw milk very low in bacteria did not

TABLE 1
Effect of concentration of methylene blue on reduction time

RATIO OF CRYSTALLINE DYE TO MILK	MILK I	MILK II	MILK III	MILK IV
	minutes	minutes	minutes	minutes
1:10,000	25	60	165	
1:20,000	25	45	165	265
1:40,000	22	42	117	240
1:100,000	15	32	95	189
1:200,000	10	27	93	163
1:300,000	8	21	77	163
1:400,000	6		57	160

reduce the dye for several days. Simmons (1919) using a concentration of 1 to 10,000 noted that some milks did not reduce the dye.

It is a well known fact that methylene blue has an antiseptic action. It seems probable that some of the workers who have used the reduction test employed such amounts of the dye that it must have retarded to a marked extent the growth of the bacteria and therefore the reduction time was prolonged.

A considerable number of trials have been made to determine the effect of the dye on the reduction time and the concentration best adapted for employment in the test. Sufficient of the data obtained to illustrate the effect of the varying concentrations on the reduction time of different samples of milk are given in

table 1. It is to be noted from the data that the reduction time is prolonged as the concentration of the dye increases, a fact that can be explained only by the injurious effect of the dye on the activities of the organism.

The tablets prepared and sold by Blauenfeldt and Tvede of Copenhagen are commonly employed in making the reduction test in the Scandinavian countries where it is most widely used. When these tablets are used according to the directions furnished therewith, the color imparted to the milk is practically identical with that obtained when 1 part of the crystalline dye is used to 200,000 parts of milk. Tests made on 24 samples of market milk show that the reduction time is practically the same with the tablets as with 1 part of the crystalline dye to 200,000 parts of milk.

Influence of different brands of dyes

Dyes from various sources, both foreign and domestic, have been compared in different concentrations. The differences in reduction time with any sample of milk, when different dyes are used, are so small as to have no practical significance.

Several writers have stressed the importance of using dyes free from impurities. It seems probable that the impurities which may be present in the grades of dyes that should be used will have little if any effect on the results in the high dilutions in which the dye is employed.

Effect of the age of the methylene blue solution

A number of writers have made the statement that the methylene blue solution to be used in the reduction test should be freshly prepared. Others emphasize the necessity of using sterile solutions of the dye. In order to determine the effect of age on solutions of the dye, tests were made with freshly prepared solutions of dyes from various sources, and with solutions which were two years old. Some of the results are presented in table 2. The results obtained with the fresh solutions and with the old

solutions are practically identical. It is, therefore, believed that the age of the solution will have little if any effect in the practical application of the test.

TABLE 2

The comparative reduction time with fresh and old solutions of methylene blue

	MARTINDALE		HARMER	
	Fresh	Two years old	Fresh	Two years old
	minutes	minutes	minutes	minutes
1:100,000	425	400	146	148
1:200,000	400	370	139	137
1:300,000	380	360	135	137
1:400,000	353	350	121	113

The effect of temperature on reduction time

Most investigators have employed 37° to 38°C. in the reduction test. It is known that some of the milk bacteria, such as the lactic,³ may grow more rapidly at slightly lower temperatures. It would, therefore, seem that a lower temperature might be more desirable in the practical application of the test. The reducing properties of various pure cultures at different temperatures have been studied by Rahn (1920) and the effect of varying temperatures on the reducing properties of market milk has been studied by Arup (1918) who believes that if milk has been stored at low temperatures, the reduction time at 30°C. will be shorter than at 38°C.

In order to gain some information concerning the reducing power of market milk at different temperatures, a number of tests were made. In each trial approximately 20 samples were employed. If the reduction time of a single sample or the average reduction time of a group of samples at 38°C. is expressed by unity, the reduction time at any other temperature may be expressed by some fraction or multiple of unity. With the great majority of samples the higher temperature has given the most rapid

³ In this paper the phrase "lactic bacteria" is used to refer to that group of organisms which is primarily concerned in the souring of milk at ordinary temperatures.

reduction. With certain samples of milk the reduction time at 28°C. has been shorter than at 38°C. There seems, however, no reason to deviate from the accustomed temperature (38°C.) in the making of reduction tests. The relative rank of a series of samples is probably of greater importance than the actual reduction time, since it is desirable that the relative quality of milk from a number of sources be determined and improvement of the supply coming from those furnishing the poorer milks be sought. In general, a series of samples of milk will be placed in the same order by the reduction test no matter what the temperature employed in the making of the test. The temperature at which the average milk will reduce most quickly is, therefore, the best one to be used.

TABLE 3

The influence of temperature on the relative average reducing power of a series of samples of milk

TRIAL	38°C.	33°C.	28°C.
1	1	1.4	2.0
2	1	1.5	1.9
3	1	1.7	2.1
4	1	1.5	2.0
5	1	1.6	1.9
6	1	1.4	2.0

Are there non-reducing bacteria?

Previous investigations claim to have found that certain bacteria would not reduce methylene blue in milk. It seems to the writers that these observations are in error. An organism which grows slowly in milk, either because of temperature, nutritive conditions, or an excessive amount of methylene blue will not be able to use oxygen as fast as it can diffuse into the milk. Such an organism would be classed as a non-reducer. Under appropriate conditions such an organism will reduce the dye. In a mixture of organisms it will exert an effect. There is every reason to believe that every organism growing in the milk assists in the reduction of the dye. All do not function in the same degree, however.

The relation of the living cell to reduction

The effect of heat on the reduction time is evidence that the cell itself and not its enzymes are the causal agents in the reduction of the dye. The data presented in table 4 show that temperatures that are not supposed to affect enzymes influence the reducing action of milk. The increase in reduction time with the increase in temperature may indicate the varying resistance of different cells of the same organism or a complexity of bacterial flora in the milk, or possibly it may indicate both conditions. The

TABLE 4

The effect of heating milk to varying temperatures on its reducing power

TEMPERATURE TO WHICH HEATED	TRIAL 1	TRIAL 2
°C.	minutes	minutes
40	17	45
45	17	50
47	17	50
50	17	55
52	32	77
54	42	107
56	57	135
58	139	155
60	189	205
62	242	240
64	377	305

effect of such a weak antiseptic as boric acid in concentration of 1 part per 2500 parts of milk approximately doubles the reduction time. The effect of increasing amounts of methylene blue in lengthening the reduction time is to be traced undoubtedly to the antiseptic action of the dye. The influence of these factors, heat and antiseptics, finds its explanation in their influence on the living cells, and it is evidence that the reduction of methylene blue is very intimately connected with the vital processes of the cell rather than with any extracellular by-products.

Effect of reaction of the milk on reduction time

Mueller (1906) studied the effect of varying reactions on the reduction time and also studied the effect of adding sodium carbonate and sodium bicarbonate to sour milk. His conclusion from rather extended observations was that the reduction time of milk in which a considerable amount of acid has been developed is not changed by neutralization of the acidity. Orla-Jensen also made similar observations. This is exactly what one would expect for it is well known that the rapidity of growth of bacteria in milk does not decrease until the acidity has reached a point at which the milk will curdle at ordinary temperatures. In other words, the reaction of any sample of milk to which the reduction test would ever be applied in practice would not be such as to interfere with the test.

Effect of shaking on the reduction time

It is recognized that one of the reasons why the plate culture does not reveal the exact number of living cells in the milk is the occurrence of clumps of organisms therein. The ordinary lactic bacteria tend to occur in twos, other forms may occur in much larger aggregates. Each aggregate, of course, gives rise to a single colony. This has led the committee of the American Public Health Association that formulated standard methods for the bacteriological examination of milk to suggest that the plate culture count of milk be spoken of as such rather than to speak of the number of bacteria per cubic centimeter. This source of error in the plate culture method should not be present in the reduction test since undoubtedly each cell of such clumps as are likely to occur in milk will function as though the other cells were not present. This should indicate that shaking would have no effect upon the reduction time. Dons (1914) found this to be true. Orla-Jensen confirmed this observation. In our own work some attention has been paid to this point. We have found that the agitation of the milk has practically no effect on its reduction time.

Relationships existing between different groups of bacteria in milk

Efforts have been made by numerous investigators to correlate the results of the plate culture method with those obtained with the methylene blue reduction test. When the correlation was not an exact one, it has had the tendency to minimize the value of the reduction test. Rahn (1920), in a recent article, has again raised this objection and considers the reduction test only an approximate method and not comparable in value to other tests.

It seems apparent that these investigators have not given adequate consideration to the relationships which must exist between the various groups of bacteria that are present in milk. Two kinds of organisms may have a favorable reaction upon each other; thus the growth of both may be stimulated. Again, one kind may be favored by the previous growth of another, or the development of one group may exert an inhibiting action upon another. An important example of this is the influence of the acid-forming group upon the liquefying bacteria. Again, inhibition of both organisms may result. When one considers that there are many groups of organisms present in milk, it is evident that the relationships existing between them must be most complex. In the plate method these relationships are avoided when the plates are not too thickly seeded. It is well known to every bacteriologist that thickly seeded plates give no correct idea of the quantitative or qualitative bacterial content of the milk. Plates heavily seeded may give the impression that nothing but lactic bacteria are present in the milk, while more thinly seeded plates from the same sample may reveal many other kinds of bacteria. The existence of these relationships between different groups of bacteria in milk and their absence in plate cultures is undoubtedly an important cause for a lack of correlation between the results obtained with the plate culture method and with the reduction test.

From many points of view the bacteriologist is chiefly interested in those organisms which are actually growing in milk. It seems probable that the methylene blue reduction test because it is influenced by these relationships measures more accurately than does any other method the bacterial activity in the milk.

Variation in reducing action of different kinds of bacteria

Another cause for the non-correlation of results obtained by the plate culture method and by the reduction test is the variation in the reducing action of different milk bacteria. Efforts have been made by a number of investigators to compare organisms with reference to their reducing power. Orla-Jensen (1912), Barthel (1908), Weigmann and Wolff (1916) and Rahn (1920) have worked along this line. The conclusions which they reached in regard to the reducing action of the different milk bacteria were quite dissimilar. Orla-Jensen and Barthel asserted that the ordinary lactic bacteria were not to be classed as strong reducing organisms, while Rahn believed them to be the most active in the reduction test. The methods used by these investigators varied. Orla-Jensen and Barthel added methylene blue to the cultures that were already fully developed, while Rahn inoculated the milk with the culture and then added the methylene blue. In the latter instance the conditions would be exactly similar to those present in the practical application of the test; namely, the organisms would be actively growing during the time of the test while, in the method used by Orla-Jensen and Barthel, only the dormant organisms would be present.

In our own work many efforts have been made to obtain some idea of the relative reducing action of different bacteria. Various methods have been employed. The one which gave the most satisfactory results was as follows: A series of tubes of fresh milk which had been heated to the boiling point were inoculated with a constantly decreasing amount of an active lactic culture. The same tubes were then treated in the reverse order with the culture of the organism to be compared with the lactic. Each tube of the series, therefore, contained the same volume of the mixed cultures of the two organisms, but the relation of the two cultures to each other was different in each tube. If the organism to be compared with the lactic had the same reducing action, and if the organisms did not influence each other unfavorably and grew at approximately the same rate, the time for reduction should be much the same in the different tubes of the series. This con-

clusion is based on the supposition that the cultures of the various organisms contained approximately the same number of cells. Although such a condition probably did not exist nevertheless the variation in number should not influence the results to such an extent as to make them of no value. Some of the results obtained are given in table 5.

It is to be noted that with each organism tested the reduction time has increased as the inoculum with lactic decreased. If the compared organism had reducing powers equal to the lactic

TABLE 5
The reducing action of different bacteria in milk

INOCULUM			COMPARED ORGANISMS												
Tube	Lactic bacteria	Compared organism	1	2	3	4	5	6	7	8	9	10	11	12	13
	cc.	cc.	min.	min.	min.	min.	min.	min.	min.	min.	min.	min.	min.	min.	min.
1	1.9	0.0	8	10	14	12	13	12	31	15	15	15	15	8	8
2	1.7	0.2	11	12	16	14	15	13	37	15	15	15	15	11	8
3	1.4	0.5	16	22	20	18	15	19	43	20	20	16	20	16	16
4	1.1	0.8	21	26	30	20	19	25	64	24	24	24	24	22	22
5	0.8	1.1	29	35	30	28	30	38	71	33	33	33	33	35	35
6	0.5	1.4	43	62	56	50	45	58	81	44	45	45	80	70	70
7	0.2	1.7	80	87	71	57	52	106	98	67	56	56	67	120	120
8	0.0	1.9	140	173	93	95	85	178	335		107	260	260	360	185

Organisms 1 to 6 inclusive were isolated from milk. They represented the udder flora. Organisms 7, 8, and 9 were representatives of the aerogenes group; 10 and 11 of the colon group; 12 and 13 of the *Bulgaricus* group.

bacteria this increase in reduction time would not have occurred for the increasing amount of the compared organism would have balanced the decrease in lactics. No organism tested was at all comparable to the lactic in reducing power. The great difference in reduction time between tubes 7 and 8 is evidence of the marked reducing action of the lactic bacteria in comparison with the other bacteria. Tube 7 contained in each trial a small amount of lactic bacteria, tube 8 none.

The reducing action is, of course, influenced by the rate of growth of the various organisms. The temperature used in the

test, namely, 38°C., is more favorable for the organisms of the colon-aerogenes group than for those of lactic group. In spite of this handicap the lactic organism exhibits much greater reducing action than do the members of the colon group. This observation agrees with what is known concerning the reducing action of these organisms in litmus milk.

The relation of reduction time to keeping quality

The desirability of a laboratory test that would give some evidence of the keeping quality of milk would be of value in grading it. In order to collect some information as to keeping quality as measured by the development of acid, and the reduction time of the milk, a number of tests were made. In the first trial tubes of fresh milk, very low in bacteria, were inoculated with sour milk in such a way that the number of bacteria added would decrease from tube to tube in any one series. The milk was stored at approximately 8°C. and after ninety-six hours the acidity of the milks was determined. The results of two such trials are presented in table 6.

It is to be noted from table 6 that the correlation between the reduction time and the development of acidity was perfect. It is to be remembered that lactic bacteria predominated in the milk used in these trials. In other words, the bacterial flora was much less complex than that of ordinary market milk.

In samples of ordinary milk not only acid-forming, but also non-acid-forming and alkali-forming bacteria will be present. There would be wide variations in rapidity of growth of the different kinds at varying temperatures. Such complex relations would probably destroy to a considerable extent the correlation between acid development and reduction time. In order to collect some information concerning this point, acid and reduction tests were run on samples of market milk when fresh, and also after storage for twenty-four and forty-eight hours, at approximately 10°C. In table 7 the samples were ranked according to their reduction time when fresh, the sample reducing most rapidly being placed first.

It is to be noted that the acidity has developed more rapidly in the samples of milk which showed a short reduction time when fresh, and in a general way there is a correlation between reduc-

TABLE 6

The relation of the keeping properties of milk to reduction time

SAMPLE	TRIAL I	
	Reduction time	Acidity after ninety-six hours at 8°C.
	minutes	
1	150	0.57
2	186	0.56
3	217	0.51
4	258	0.44
5	303	0.33
6	340	0.28
7	355	0.24
8	371	0.22
9	403	0.20
10	411	0.19

TABLE 7

The relation of acidity and reduction time in milks after varying periods of storage

SAMPLE	ACIDITY AND REDUCTION TIME AFTER 0 HOURS AT 10°C.		ACIDITY AND REDUCTION TIME AFTER 24 HOURS AT 10°C.		ACIDITY AND REDUCTION TIME AFTER 48 HOURS AT 10°C.	
	per cent	minutes	per cent	minutes	per cent	minutes
56	0.222	125	0.261	10	0.344	1½
30	0.166	205	0.205	11	0.355	2
369	0.188	305	0.194	55	0.210	7
59	0.188	340	0.194	265	0.205	11
86	0.188	360	0.200	260	0.200	22
130	0.210	360	0.210	295	0.216	22
354	0.216	400	0.228	49	0.235	5
135	0.166	400	0.183	335	0.177	160
356	0.188	475	0.188	335	0.177	75
359	0.194	503	0.188	355	0.177	90
357	0.194	510	0.200	385	0.222	15

tion time and keeping quality if the latter is to be measured by acid production. There are, however, certain deviations from this correlation; for example, sample number 135 has shown practically no increase in acid during the storage period, while

the reduction time has decreased. The same is true of samples 356 and 359. In other words, in these samples the effect of acid-forming bacteria if they were present, has been overbalanced by alkali-forming organisms. The latter would, of course, exert an effect on the reduction time. They would not, however, influence the keeping quality as measured by acid development.

It seems from the results obtained that the reduction test will reflect only in a very general way the keeping quality of a sample of milk. It is undoubtedly true that the other methods for the examination of milk, such as the plate culture method and the direct microscopic examination indicate quite as well as the reduction test, the keeping quality. On account of the great complexity of bacterial flora in milk, it seems impossible for the keeping quality to be determined by any one test, or probably by any group of tests.

Directions for making the reduction test

A stock solution of medicinal methylene blue or that recommended for use in staining bacteria is prepared by dissolving one part of the crystalline dye in 2000 parts of water. A portion of this solution is further diluted shortly before use until the concentration is one of the dye to 20,000 of water. One cubic centimeter of this solution in 10 cc. of milk gives a dilution of slightly over 200,000. The solution should not be filtered since this will remove a considerable portion of the dye, especially is this true with dilute solutions.

Tubes of approximately the same diameter and of such a size that 11 cc. will not fill them over one-half full are used. The samples should be collected with the same care as for any bacteriological examination. The tubes should be clean and steamed or boiled shortly before using. It is unnecessary to sterilize them. The 10-cc. sampling pipette should be clean and should be rinsed at least three times with boiled water between samples. After the addition of the dye the contents of the tube should be mixed by closing the tube with the thumb or palm and inverting once or twice. Wiping the moistened area of the hand with a clean

towel will be sufficient prevention of contamination from sample to sample.

A water bath which can be kept at 38°C. in which the tubes can be placed is the only apparatus needed, other than the tubes and pipettes.

The frequency of observation will be determined by the number of grades into which one wishes to divide the milks. There will be little use under ordinary conditions of extending the period of observation over six hours.

Reading the test

It is usually thought that the distribution of bacteria in a recently mixed sample of milk is uniform, in other words, that any small quantity will be an exact duplicate of another sample. There are a number of reasons for believing that this is not true: especially in milk of low bacterial content, among them is the appearance of some of the milk in the reduction test. In the great majority of cases, the color will disappear uniformly from all parts of the milk. In other cases the color may disappear in an uneven way. A small zone or area may lose the color before there is any apparent reduction in other zones. In still other cases the color may persist in the lower layers of the tube after the upper part is completely decolorized. The more common occurrence is the persistence of the color at the top. Vitoux (1920) in his description of the reduction method as used in Holland states that the upper one-fourth of the tube should be disregarded in reading the test. Another method, less convenient in practice, is to take as the end point, the time when no blue color is to be noted after the milk has been mixed. The slight amount of unreduced dye in the surface layer will not be noticeable when mixed with the entire quantity of milk. Again, the color may be reduced to such a point that the milk appears white except when compared with a control tube of milk when a distinct bluish tinge will be noted. This residue of color may persist unchanged for a considerable period. We have not been able to relate this to any factor. It is probable that it is of small importance.

SUMMARY

The importance of the bacterial content of milk in determining its quality is discussed and the need of some simple and inexpensive method for the bacteriological analysis of milk is presented. The methylene blue reduction test seems to meet the need.

The literature treating of the reduction test has been reviewed and a study has been made of a number of factors which influence the results obtained with the test. In the work herein reported attention has been directed to the influence of the concentration of the dye, to the age of the solution of the dye, and to the source or brand of the methylene blue.

The influence of temperature on the reduction time has been investigated.

The direct dependence of the test on the vital activities of the cells is shown by the effect of heating the milk to relatively low temperatures and by the effect of antiseptics.

The relationships which may exist between different groups of bacteria are discussed and data are presented to illustrate the effect of these relationships in influencing the reduction time.

The reducing action of different groups of milk bacteria has been studied as well as the relation of reduction time to keeping quality.

Directions for making the test are given.

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THE COPPER CONTENT OF COWS' MILK

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A review of the literature will reveal many records of the presence of traces of common metals in animal and vegetable substances, although little or no attention is given to these elements in the majority of the published analyses. Obviously, the circumstances governing the analysis of such substances rarely warrants qualitative or quantitative determinations for these minute amounts of metal. This is especially true because, with the exception of iron, it is not known whether their presence is merely accidental or whether they are necessary prerequisites for certain functional activities.

Zinc and copper have been reported in measurable amounts in numerous food substances, and by correlating the results obtained by different investigators it is not difficult to account for their presence in the animal organism. There appears to be authentic data from which it is possible to trace these elements—particularly copper—back to foods of vegetable origin and even to the soil upon which the plants were grown. From the examination of 140 samples of soil Maquenne and DeMoussy (1) found amounts of copper varying from 1 to 250 mgm. per kilogram of earth. They also report copper as a normal constituent of many edible plants, and furthermore, it is stated that this metal is present in all parts of the plant. In many instances the amount of copper was found to greatly exceed that of the soil in which the plant was grown. Guerithault and Maquenne (2), using a somewhat different method of analysis but one for which a high degree of accuracy is claimed, state that copper is found regularly in many digestible substances of vegetable origin. The following table compiled from their data shows a wide range of vegetable products in which copper was found in significant amounts.

Fleurent and Levi (3) have extended these investigations to other vegetable and animal substances. For milk they report 1.4 mgm. of copper per liter; for egg yolk 20 mgm. per kilogram of dry substance; and for horse flesh 22 mgm. per kilogram of dry substance. Rost (4) reports the presence of zinc and copper in measureable amounts in the flesh and organs of various animals, in milk and blood, and in human excreta. Zinc and copper are

TABLE 1
Copper content of digestible substances of vegetable origin
(Guerithault and Maquenne)

MATERIAL	Cu PER 100 GRAMS ASH	Cu PER KILOGRAM OF DRY MATERIAL	Cu PER KILOGRAM OF FRESH MATERIAL
	mgm.	mgm.	mgm.
Sorrel (leaves).....	20.7	27.5	2.4
Spinach (leaves).....	9.0	18.3	1.8
Lettuce (leaves).....	18.0	36.3	2.0
Carrots (roots).....	18.0	17.6	2.2
Turnips (roots).....	36.0	29.7	3.0
Radishes (roots).....	33.0	52.7	3.8
Mangels (roots).....	26.6	24.5	3.2
Potatoes (tubers).....	15.0	7.7	1.9
Pumpkins.....	12.0	10.9	1.1
Apples.....	11.5	7.4	1.2
Pears.....	19.6	12.9	2.2
Beans.....	25.4		10.0
Peas.....	16.9		7.2
Soy Beans.....	17.3		9.0
Wheat.....	18.0		7.2
Rye.....	19.3		7.5
Barley.....	14.6		6.5
Oats.....	22.2		17.1
Maize.....	12.3		6.8

commonly associated together in these substances, and from the data presented zinc was usually found in greater amounts than the copper. It is concluded that these elements are ingested with the food, absorbed from the gastro-intestinal tract, stored in the muscles and in the liver and finally pass into the body excretions and secretions. Thudichum (5) was the first to report the presence of copper in the human brain. Recently Bodansky

has substantiated his observations by quantitative determinations of the copper in the brains of four adults and one foetus five months old. The amount of copper per kilogram of adult brain was found to vary from 3.6 to 6 mgm., whereas the amount found in the fetal brain was 6.8 mgm. The presence of copper and zinc in the tissues of certain marine organisms has been known for a long time and the recent works of Rose and Bodansky (7), and Hiltner and Wichman (8) have furnished further data relative to the copper content of these organisms. Data recently published by Birekner (9) showing the zinc content of many plant and animal substances, but particularly milk, seems to leave no doubt but what this metal is present in measurable quantities in many common foods; determinations for copper were not included in this investigation. Bertrand (10), quoting the work of several investigators states that meat and milk contain 0.50 mgm. of copper per kilogram. (The original article to which reference is made has not been found by us.)

EXPERIMENTAL

Although references to the presence of copper in cows' milk have been found in the literature, the data are meager. The purpose of this paper therefore, is to furnish data showing the variations in copper content of cows' milk, freshly drawn, and after exposure to metallic copper as might occur in milk-handling establishments. The data have been collected over a period of two years in connection with researches on desiccated milk products.

The estimation of copper in milk

The principle difficulty encountered in determining the amount or even the presence of minute traces of copper in milk lies in the selection of method which is sufficiently delicate to detect slight variations without resorting to the use of large quantities of material. Many of the methods commonly used for the determination of copper in foods, while reliable in their specificity for copper and in their quantitative measurement of as low as 1 to 5 mgm., are of no value for indicating amounts as low as 0.1 or

0.01 mgm. Some of these methods however, were used as preliminary tests upon large quantities of freshly drawn milk or milk powder known to be uncontaminated by copper.

The method described in detail by Rose and Bodansky (7) with suitable modifications for liquid milk, yielded sufficient copper sulphide for the positive identification of copper in freshly drawn milk. Qualitative tests for copper on the concentrated liquid from which no more sulphide could be precipitated showed that slight traces of copper were still present. Therefore while the method served to indicate the presence of copper in natural milk, it was not considered sufficiently delicate for the correct measurement of the small amount present.

The potassium ferro-cyanide method was likewise used as a preliminary test for the positive identification of copper in freshly drawn milk. The method was applied to milk ash from which the iron had been removed, and in the presence of traces of zinc salts. A distinct pink coloration was formed indicating the formation of cupric ferro-cyanide. After a period of time the pink color disappeared with the formation of a small amount of dark brown precipitate. In view of the observations of Maquenne and Demoussy (11) who claimed that this reaction is sufficiently sensitive to detect from 1 to 1.5 mgm. of copper per liter, it was considered that these results furnished further confirmatory evidence of the presence of traces of copper in uncontaminated cows' milk.

As further evidence of the presence of copper in milk, 200 grams of milk powder (equivalent to approximately two liters of liquid milk) known to be uncontaminated by copper were examined by the tentative method for determining copper in foods as described in "Methods of analysis of the Association of Official Agricultural Chemists" (12). Since this method is dependent upon the precipitation of sulphide, preliminary tests were made to determine its effectiveness in measuring small amounts of copper in solutions of known copper content. The results of these tests show that the method was unsatisfactory for the correct measurement of copper in quantities less than 1 mgm. From the 200-gram sample of milk powder, sufficient copper sulphide was precipitated however, to permit the identification of the metal.

The method which was found to be most satisfactory for such small amounts of copper as are found in milk, and one which at least gives comparative results without excessively tedious manipulations, is the potassium ethyl xanthate method described by Scott and Derby (13). The method is especially recommended for detecting traces of copper in salts crystalized in copper containers. In addition to its high degree of sensitiveness, it has the advantage that small quantities of iron, lead, nickel, cobalt, zinc and manganese do not interfere. The details of the method as applied to milk follow:

The method is based upon the fact that small amounts of copper react with potassium ethyl xanthate to produce a yellow color. The intensity of the yellow color is in direct proportion of the amount of copper present. When large amounts of copper are added to the xanthate reagent a yellow precipitate of copper xanthate is produced.

Stock solution of copper sulphate: 3.9283 grams of $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ are dissolved in water and made up to 1000 cc. One cubic centimeter is equivalent to 0.001 gram of copper.

Standard copper sulphate solution: 10 cc. of the stock solution are diluted to 1000 cc. with distilled water. One cubic centimeter is equivalent to 0.00001 gram of copper.

Potassium ethyl xanthate solution: One gram of potassium ethyl xanthate is dissolved in 1000 cc. of water. This solution should be stored in an amber colored glass stoppered bottle.

Method: 50 to 100 cc. of milk are evaporated to dryness in a silica or porcelain evaporating dish and ignited with a low red heat until a white ash is obtained. Dissolve the ash in the least possible amount of concentrated hydrochloric acid, add about 10 cc. of copper-free distilled water, heat to boiling and filter. If a residue remains on the filter it should be ashed, dissolved in concentrated hydrochloric acid and added to the first filtrate. To the dissolved ash add an excess of concentrated ammonium hydroxide and filter. Wash the precipitate with distilled water. The precipitate is then dissolved in warm dilute hydrochloric acid and re-precipitated with ammonium hydroxide, again filter and wash. The combined filtrates are boiled to drive off excess ammonia. When no more ammonia can be detected, cool, add one drop of concentrated hydrochloric acid followed by 2.5 cc. of 0.5 N sodium hydroxide, and 8 cc. of 0.1 N hydrochloric acid. The solution should be

of neutral reaction at this point, but under no circumstances must it be alkaline. Merely driving off the excess ammonia leaves a slightly alkaline solution which if added to the xanthate reagent causes low results due to the gradual destruction of the color formed by the copper.

The solution containing copper, treated as described above is poured into a 100-cc. Nessler tube, 10 cc. of the potassium ethyl xanthate reagent added, and water added to bring the volume up to the mark. To a series of similar tubes containing 50 to 60 cc. of water add 10 cc. of the xanthate reagent. Then from a burette add to the first tube 0.5 cc. of the standard copper sulphate solution; to the second tube 1 cc. to the third tube 1.5 cc., etc. Add sufficient copper-free water to each tube to bring up to the mark. Compare the tube containing the unknown with the tubes containing the known amounts of copper. If the color of the unknown solution is between two of the standards, gradually add more of the standard copper sulphate solution to the tube containing the lower amount of this standard solution. For example, if the color of the unknown sample falls between the 1-cc. and 1.5-cc. standards add the standard copper solution a drop at a time to the 1-cc. tube until the color in the tube containing the unknown and in the tube to which more of the standard has been added match in intensity. Record the total amount of standard copper sulphate solution used. From this figure the percentage, milligrams per liter, or parts per million of copper in the original sample can easily be determined.

It is necessary to test all reagents for copper and if detectable amounts are found blanks should be run and proper corrections made.

The reliability of the potassium ethyl xanthate method can be shown best by the figures in the following tables. In table 2 is shown the copper found in 10 cc. volumes of water to which known amounts of copper had been added. Samples containing the different amounts of the metal were analyzed by two different analysts who had no knowledge whatsoever of the amounts of copper present. In table 3 is shown the amount of copper recovered from a sample of milk to which definite amounts of the metal in the form of a common copper salt had been added. The amount of copper present in the original sample was known to the analyst but he did not know the amount of metal which had been added. From the results of these determinations it appears

that the method can be considered fairly reliable for copper in milk to as low as 0.005 mgm. Since this amount is considerably less than will be shown to have been present in the samples examined, it is reasonable to assume that the results hereinafter recorded have not been appreciably affected by inadequacies of the method used.

Amount of copper in freshly drawn cows' milk

In order to determine the copper content of milk drawn directly from the cow, several samples from different animals in different

TABLE 2*

Copper in aqueous solution as determined by the potassium ethyl xanthate method

SAMPLE	AMOUNT OF COPPER PRESENT	AMOUNT OF COPPER FOUND (NO. 1)	AMOUNT OF COPPER FOUND (NO. 2)
	mgm.	mgm.	mgm.
1	10	10	Not determined
2	5	5	5
3	1	1	1
4	0.5	0.5	Not determined
5	0.1	0.1	0.09
6	0.02	0.02	0.015
7	0.002	0.002	Not determined
8	0.001	0.001	0.001
9	0.0005	Trace or none	None

TABLE 3*

Copper in milk as determined by the potassium ethyl xanthate method

SAMPLE	AMOUNT OF COPPER IN 100 GRAMS MILK	AMOUNT OF COPPER ADDED	TOTAL AMOUNT OF COPPER PRESENT	AMOUNT OF COPPER FOUND
	mgm.	mgm.	mgm.	mgm.
1	0.04	0.02	0.06	0.065
2	0.04	0.005	0.045	0.050
3	0.04	0.005	0.045	0.045
4	0.04	0.004	0.044	0.044
5	0.04	0.003	0.043	0.040
6	0.04	0.002	0.042	0.045
7	0.04	0.002	0.042	0.040
8	0.04	0.001	0.041	0.040
9	0.04	0.001	0.041	0.045
10	0.04	0.0005	0.0405	0.040

* Indebtedness to Mr. D. I. Stadden of the Laboratory Staff for part of the analyses shown in these tables is hereby acknowledged.

herds were milked directly into the glass containers and at no time were they allowed to come in contact with metal vessels.

TABLE 4
Copper content of freshly drawn milk

SAMPLE	HERD	COW	COPPER PER LITER OF MILK	RATION
			<i>mgm.</i>	
1	I	1	0.75	All cows in herd I received corn, oats, oat straw, and corn stover
2	I	2	0.65	
3	I	3	0.65	
4	I	4	0.45	
5	II	1	0.50	All cows in herd II received corn- meal, wheat bran, oats, ensilage, and hay
6	II	2	0.45	
7	II	3	0.60	
8	II	4	0.55	
9	III	1	0.55	All cows in herd III received oats, gluten, cotton seed meal, and ensilage
10	III	2	0.60	
11	III	3	0.70	
12	III	4	0.80	
13	IV	1	0.30	Ration not recorded
14	IV	2	0.60	
15	IV	3	0.55	
16	V	1	0.60	Ration not recorded
17	V	2	0.20	
18	V	3	0.30	
19	VI	1	0.40	Ration not recorded
20	VI	2	0.50	
21	VI	3	0.30	
22	VII	Mixed sample from 5 cows	0.60	Fresh grass only
23	VII	Mixed sample from same 5 cows	0.40	Highly concentrated with straw as roughage
Average all samples.....			0.521	

The ration being fed at the time the samples were taken was noted whenever it was possible to obtain this informaton. The

results obtained from these determinations are shown in table 4 from which it is evident that a small amount of copper is a regular constituent of uncontaminated cows' milk.

Other analyses of aliquot samples of milk received at milk-receiving plants which buy directly from farmers, and which represented the milk from several hundred cows were made at different times for a period of one year. Of 22 such samples analyzed the maximum copper content was found to be 0.6 mgm. per liter; the minimum 0.4 mgm. per liter; and the average 0.45 mgm. per liter. There was no consistent difference in the copper content of milk produced during the season of pasture feeding as contrasted with the milk produced during the season of stall feeding. It is believed that these results taken in conjunction with those obtained from individual cows represent averages which are applicable to all normal milk. In interpreting these results however, it is to be remembered that larger amounts of copper may be found occasionally in milk which has been exposed to copper utensils during handling and processing before it reaches the consumer.

Copper taken up by milk during handling in milk plants

Several series of analyses have been made in order to determine the amount of copper taken up by milk under conditions to which it might be subjected in passing through various processes commonly employed in milk-handling establishments. Table 5 shows the results obtained in a factory equipped throughout with the common sanitary type milk pipe (tin-lined copper pipe) and with glass enameled tanks for storage purposes or for use in pasteurizing. The entire volume of milk received passed through about 200 feet of sanitary pipe, which upon close inspection was found to have but a very slight amount of exposed copper at a few places where the tin had been worn from the pipe. In its journey through the pipe lines the milk passed several bronze or brass valves, tees and unions very few of which were completely covered with tin. From the time the milk entered the factory until it was ready to leave the only opportunity for taking up

copper was during its passage through the valves, tees, etc., to which reference has just been made. The results given in table 5 show that under the conditions just described little or no copper was taken up by the milk while in the factory. Similar results are to be expected in other plants where equipment is kept in a similar condition.

In contrast with the results obtained where the minimum copper surface is exposed to the milk are those which were found on occasions where the copper content had increased from the average figures of 0.4 to 0.5 mgm. per liter up to as high as 2 mgm. per liter, for no apparent reason other than as a result of an appreciable area of exposed copper in the pipes from which the tin had been worn off. While the maximum figure of 2 mgm.

TABLE 5

Copper content of milk after passage through a well equipped factory
(Results expressed as milligrams per liter)

SERIES	COPPER IN MILK UPON ARRIVAL AT FACTORY	COPPER IN MILK AFTER PASSING THROUGH FACTORY	AMOUNT OF COPPER TAKEN UP IN FACTORY
	mgm.	mgm.	mgm.
I	0.60	0.75	0.15
II	0.45	0.45	None
III	0.60	0.60	None
IV	0.45	0.45	None

per liter was found in milk after exposure to the conditions just referred to, there were numerous samples which showed a much smaller amount of copper taken up after passage through pipes which could not be considered in a perfect condition, but which were worn to a greater extent than those from which the results of table 5 were obtained. Even though it is apparent that sanitary pipes from which tin has been worn off are a source of added copper in milk, the amount taken up in this manner is believed to be slight as compared with that which may be taken up under other conditions. Even with sanitary pipes which appear to be adequately covered with tin, a very important source of added copper has been found to be the accumulation of soluble copper salts in crevices and on rough surfaces where the bronze

or brass unions, tees, etc., are joined to the tin-covered pipes; the ground surfaces of the valves may under certain conditions become an important source of such material. Obviously, milk passing over these accumulations of soluble copper compounds will take up more copper than by passing over the clean metal of an unprotected copper pipe. Strict sanitary measures must be

TABLE 6

Copper taken up by milk by storing or heating in contact with the metal

SAMPLE	TREATMENT	AMOUNT OF COPPER ORIGINALLY IN MILK	AMOUNT OF COPPER IN MILK AFTER TREATMENT	AMOUNT OF COPPER TAKEN UP BY MILK
		<i>mgm. per liter</i>	<i>mgm. per liter</i>	<i>mgm. per liter</i>
1	Held in contact with copper pipe twelve hours at 45°F.....	0.60	0.96	0.36
2	Heated to 150°F. for two hours in contact with copper pipe.....	0.60	5.00	4.40
3	Held in contact with copper pipe twelve hours at 45°F.....	0.70	1.00	0.30
4	Heated to 150°F. for two hours in contact with copper pipe.....	0.70	2.50	1.80
5	Held in contact with copper pipe twelve hours at 45°F.....	2.00	3.00	1.00
6	Heated to 150°F. for two hours in contact with copper pipe.....	2.00	4.40	2.40
7	Held in contact with copper pipe twelve hours at 45°F.....	1.00	3.20	2.20
8	Heated to 150°F. for two hours in contact with copper pipe.....	1.00	5.00	4.00
9	Held in contact with copper pipe twelve hours at 45°F.....	0.60	2.80	2.20
10	Heated to 150°F. for two hours in contact with copper pipe.....	0.60	8.00	7.40

used regularly to prevent the accumulation of copper compounds in such places.

The copper content of milk as affected by condensing in copper vacuum pans has been shown to be measurably increased, but not to the extent which might be expected in view of the intimate contact between the milk and the copper. Samples of milk which had been condensed at a temperature of 110° to 120°F.,

and which were in contact with the copper condensing pan for a period of one and one-half to two hours were diluted back to the same concentration as the original milk and the copper content determined. The milligrams of copper per liter found in each of six samples follows: No. 1, 3.5 mgm.; no. 2, 1.0 mgm.; no. 3, 0.9 mgm.; no. 4, 1.3 mgm.; no. 5, 1.1 mgm.; no. 6, 3.5 mgm. Considering 0.52 mgm. of copper per liter as the average amount found in normal milk, the increase in the condensed milk samples has varied from 0.38 mgm. to 2.98 mgm. per liter.

The heating or storing of milk in copper containers or in contact with metallic copper materially increases its copper content. The results shown in table 6 illustrate the extent to which this metal may be taken up by milk under the conditions indicated. In each instance the area of copper and the volume of the milk were the same. The average amount of copper taken up by milk after holding for twelve hours at a low temperature was 1.21 mgm. per liter; the average amount taken up after heating for two hours at 150°F. was 4 mgm. per liter.

SUMMARY

Copper was found to be a normal constituent of freshly drawn cows' milk. The amounts found in the milk of individual animals varied from 0.2 to 0.8 mgm. per liter. The average amount found in 23 such samples was 0.52 mgm. per liter. On the basis of results from several samples representing mixed milk taken during periods of pasture feeding and stall feeding, there appears to be no difference in the copper content of milk as affected by the two different types of ration. The maximum amount found in these samples was 0.6 mgm. per liter and the minimum was 0.4 mgm. per liter.

The potassium ethyl xanthate method proved to be the most satisfactory method for the quantitative measurement of the small amounts of copper found in normal milk. Tests to determine the reliability and delicacy of this method show that as low as 0.005 mgm. could be measured with a fair degree of accuracy. Other methods however, were used to advantage for the qualitative detection of copper in large quantities of milk.

The amount of copper in milk may be measurably increased by storing or heating in the presence of metallic copper. Slight increases in the copper content of milk may also result from the passage of milk through the sanitary type copper pipes from which the tin has been worn off. Copper taken up from this source however, is not as great as would result from the formation of copper compounds around brass or bronze fittings as a consequence of poor sanitary practices.

The significance of minute amounts of copper in plant and animal tissues is as yet unknown. Its presence in milk, particularly when taken up as extraneous contamination may prove to be significant in connection with the high susceptibility of the antiscorbutic vitamine to oxidation. In fact, Hess (14) has already reported the destruction of this vitamine by accelerated oxidation as a result of pasteurizing milk in a copper vessel.

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METHODS OF MEASURING THE VOLUME OF CREAM ON MILK

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In the city trade the appearance and volume of the cream on the milk as delivered to the consumer is a matter of considerable commercial importance. This has long been recognized by the milk industry and different methods of measuring the cream have gradually grown up in the different plants. There is, however, an almost complete lack of literature on this subject.¹ The present publication records an attempt to bring together and study these various methods. Finding that they were not satisfactory for the study of the influence of plant operations on the volume of cream, there has been developed a simple and accurate method of measurement which has been found readily applicable under working conditions in commercial milk plants.

METHODS OF MEASURING THE VOLUME OF CREAM

The simplest method of estimating the volume of cream and probably the one earliest used consists in standing bottles beside each other and comparing the depth of the cream layers. Variations are discernible in this way provided the two bottles are of the same size and shape. While there is much to be said in favor of this method it is too crude to be of much service in determining the exact volume of the cream or in tracing the factors which combine in producing the final cream layer which attracts or disappoints the prospective customer.

¹ A more extended discussion of this matter may be found in Ill. Agr. Exp. Sta. Circular 249, 1921.

Measuring from the top of the bottle

A simple test and one much used by dealers is to measure the distance from the top of the bottle to the line dividing the cream from the milk. This was the test applied by Farrington and Russell (1) in their original study of the applicability of 140°F. to the pasteurization of milk. Such measurements do not give the true depth of the cream layer since the bottom of the bottle cap is approximately $\frac{1}{4}$ inch below the top of the bottle.

Measurements of the depth of the cream layer in the neck of bottles are comparable when made in bottles of the same size and shape and with equal depressions for the bottle cap. While it would seem a simple matter for any milk company to keep a supply of uniform bottles, such is not the case. Comparison between measurements in the bottles of different companies is further complicated by the fact that some companies are using narrower and taller bottles which increase the depth of the cream layer.

Marking the cream line and determining the volume

Where measurements must be made of the cream which has already risen in the bottle probably the most accurate available method is to mark the line between the cream and the milk and determine the volume of each. If the bottle is dry on the outside the cream line may be easily marked with a colored wax pencil. A finer line may be made with a brush and ink. Where a brush is not at hand or where the bottle is moist a file may be used.

The magnitude of the error in thus determining the amount of cream present may be tested by repeatedly measuring the volume above the mark in the same bottle. The results of ten such successive measurements in each of a dozen milk bottles where the cream line was marked by a fine ink line showed variations in the determinations of each bottle ranging from 4 to 12 cc. These same variations, expressed in terms of cream percentage, range from 0.4 to 1 per cent, with an average variation of 0.7 per cent.

Measuring the depth of the cream layer in glass tubes

It has long been the custom in making laboratory tests of the creaming ability of milk to put the milk to be tested into graduated cylinders. Cylinders with a capacity of 100 cc. have been most frequently used. While this method is well suited to the laboratory study of a few samples it is not well adapted to handling a large number of samples in a milk plant. Not only are such glass cylinders awkward to transport and store during the period in which the cream is rising but the first cost is heavy and the breakage considerable.

Hammer and Hauser (2) modified this plan in their studies by using large test tubes (size not stated) which they filled with milk to a depth of 6 inches. They measured the depth of the resulting cream layer in sixteenths of an inch and recorded it on a percentage basis, the depth of the cream layer being compared with the depth of the cream layer on a control sample of raw milk. The use of percentages in expressing the results of these studies was unsatisfactory largely because these percentages were based upon the creaming ability of raw milk.

In his later studies of the creaming ability of milk, Hammer (3) used Nessler tubes filled to a line 9 inches from the bottom. The unit used in recording the depth of the cream layer was the sixteenth of an inch and the readings were recorded to the nearest half unit. The Nessler tubes were accurately made and in many ways well adapted to these tests. While more easily stored and handled than graduated cylinders, like them, they are quite expensive and under the conditions which must almost necessarily accompany the taking of large numbers of samples are easily broken.

THE DEVELOPMENT OF A METHOD OF MEASUREMENT SUITED
TO MILK PLANT CONDITIONS

A successful study of the factors affecting the cream layer on a bottle of milk must be conducted largely at the milk plant. A workable method for studying these factors should combine simplicity (so as to permit the taking of a large number of samples) with a high degree of accuracy.

The first step in the present study was the selection of the sample tube. Hammer and Hauser used a large test tube, filled to a depth of 6 inches. This was abandoned by Hammer because of the shortness of the milk column and the consequent lack of sensitiveness of the measurements. There was selected for the present purpose a thick walled, glass, test tube, without lip, 1 inch in diameter and 10 inches long. This is a stock size and therefore not unduly expensive.

The next step was to decide upon the depth of the milk sample. Through a desire to have this comparable with the depth of the milk in the quart bottle, it was arbitrarily placed at 216 mm., approximately $8\frac{1}{2}$ inches. However, as pointed out later, there are decided advantages in choosing a slightly different length.

The desired length is marked on each tube by the use of a suitable gage and a sharp file. A tin tube, slightly larger than the glass tube and of the desired length makes a good gage. The glass tube to be marked is inserted in the tin tube, the file is held against it at the edge of the tin tube, and the glass tube is rotated slightly. In this way the glass tube can be quickly and accurately marked. The danger of the glass tube breaking at the file mark can be practically obviated by keeping the file moistened with turpentine during the marking process. In using these tubes to measure the creaming power of the milk before and after any treatment, it is well to fill three or more tubes from the same sample. Using the average of these readings increases the accuracy of the determination. Where the milk to be tested is well mixed and the tubes are filled quickly from a common sample, the resulting cream layer rarely varies more than 1 mm. in depth.

The temperature at which the samples are held is important. The studies of Hammer agree with commercial experience in suggesting that the temperature at which milk is held is a factor affecting the depth of the cream layer. Accordingly, samples to be compared should be kept at the same temperature.

In cold weather the milk at the receiving vat may be at 34°F., while that in the pasteurizer may be at 147°F. To bring samples to the same temperature, it is well to put the tubes at once into

ice water. The samples quickly take this temperature, and they may then be removed to the bottle storage room, which has a temperature of about 40°F. It would undoubtedly result in a deeper layer of cream in the tubes and more uniform results if the samples could be kept at lower and more constant temperatures. However, where the results from the tubes are to be compared with those from the bottles, there are advantages in holding both at the same temperatures.

CALCULATION OF THE VOLUME OF ROUND BOTTOMED TUBES

One of the advantages of the Nessler tube used by Hammer was the fact that it had a flat bottom. When this flat-bottomed tube was filled with milk and later the cream layer was measured, dividing the depth of the cream layer by the depth of the milk gave the percentage of cream by volume.

With a round-bottomed tube, the percentage of cream cannot be figured in quite the same way because the rounded portion of the tube does not contain as much milk as would a flat-bottomed tube of the same length. A necessary step is to determine the length of a cylinder having the equivalent volume.

In the case of the tubes graduated at 216 mm. the following method was used. Starting at the 216 mm. mark, a distance of 200 mm. toward the bottom of the tube was indicated by a fine ink line. The tube was successively filled to the two graduations and the volumes noted, standardized burettes being used.

The computation of the length of the cylinder having the same volume as the lower 16 mm. of the tube was made according to the following simple proportion:

Volume of 200-mm. portion : 200 :: volume of 16-mm. portion : x , where x is the length of the equivalent cylinder.

In connection with studies of the creaming power of milk at various milk plants, separate consignments of tubes of these dimensions were purchased through the regular commercial channels. Sixty-two tubes were selected as representative from four such shipments, and their volumes were determined according to the above method. These measurements showed that these 62 round-bottomed tubes measuring 216 mm. in length

had, on the average, the same capacity as flat-bottomed tubes measuring 212 mm. (The extremes of variation were equal to tubes 1.3 mm. longer and 1.1 mm. shorter than the average.)

When the tubes are filled with milk, there ordinarily develops a cream layer of not more than 30 mm.; this gives a ratio of 1 mm. of cream to about 7 mm. of total length. Under such circumstances the above slight variation in the length of the different tubes is not sufficient to measurably affect the final reading. Accordingly, the volume of these tubes at a depth of 216 mm. may be taken as equivalent to that of a flat-bottomed tube 212 mm. long.

The object of these measurements of the tubes was to provide a basis for converting the measurement of the cream layer, given in millimeters, into percentage of cream by volume. These readings may be thus transformed by dividing the reading by 212, or by multiplying it by 0.47. The conversion of the measurement of the depth of the cream layer into percentage by volume can be even more easily done if the length of the sample tube is shortened by 12 mm., so that each millimeter in depth of cream is equivalent to 0.50 per cent by volume. In this case the tube should be filled to a depth of 204 mm., or 8 inches.

EFFECT OF DIAMETER OF TUBE ON CREAM LAYER

The Nessler tube suggested by Hammer has a diameter of about $\frac{1}{4}$ inch smaller than the one here proposed. The question whether this variation in diameter would have any measurable effect upon the depth of the resulting cream layer was tested in the following manner.

There were prepared 50 of the 1-inch, round-bottomed tubes calibrated at a depth of 216 mm., and a like number of the flat-bottomed Nessler tubes calibrated at a depth of 212 mm. Twenty-five tubes from each set were filled with raw milk, as delivered from the barn, at a temperature above 90°F.; and the remaining 25 tubes from each set were filled with milk just pasteurized at 142°F. for thirty minutes. These tests were repeated on two successive days. The tubes of milk were immediately placed in ice water, and when cool were transferred

to a cooler kept at about 40°F. The depth of the cream layer was recorded in millimeters at the end of twenty-four hours.

The average depth of the cream layer developing on the raw milk on the first day was 28.90 mm. in the 1-inch tubes and 28.94 mm. in the Nessler tubes. The corresponding depth of cream rising on the pasteurized milk were 31.20 and 31.22 mm. These averages are as close as could be expected from an average of two sets of the same tubes. On the second day the cream layer in the 25 1-inch tubes of raw milk averaged 33.70 mm. and in the Nessler tubes, 33.14 mm. The corresponding measurements with pasteurized milk were 31 mm. and 30.90 mm. If the fifty determinations with each kind of tube were averaged, the result would be 30.05 mm. with the 1-inch tubes and 30.09 with the Nessler tubes. The results make it evident that when cylinders differing in diameter from $\frac{3}{4}$ inch to 1 inch are filled with milk to the same depth and held at the same temperature, they will later develop equal layers of cream.

COMPARISON OF VARIOUS METHODS OF MEASURING THE CREAMING ABILITY OF MILK

Attention has already been called to the various methods now in use in milk plants for measuring the volume of cream. A comparison of the results obtained by these different methods is made possible by applying them simultaneously to the same milk. Accordingly bottles were taken from the bottler and immediately placed in the cooler for about twenty hours. The depths of the cream layers were then measured, the cream line was marked, and the volume of the cream and the per cent of cream in each bottle determined. When these bottles were selected at the bottler additional bottles were taken and from them test tubes were filled, the tubes of milk cooled in ice water, and placed in the cooler with the bottles for about twenty hours. The depth of the cream in these tubes and the per cent of cream which these readings represented was also noted. The results of these comparable observations are given in table 1.

From the data in table 1 it is evident that the measurement from the top of the bottle is an inaccurate method of determining

the amount of cream present. Among the nine bottles measuring three inches of cream the per cent by volume present ranged

TABLE 1
Comparison of various methods of cream measurement

SAMPLE NUMBER	MEASURED IN THE BOTTLE				MEASURED IN TUBE	
	Cream depth	Bottle volume	Cream volume	Cream	Cream depth	Cream
	<i>inches</i>	<i>cc.</i>	<i>cc.</i>	<i>per cent</i>	<i>mm.</i>	<i>per cent</i>
1	2 $\frac{1}{8}$	938	98	10.45	22.0	10.38
2	2 $\frac{1}{8}$	940	103	10.96	23.5	11.09
3	2 $\frac{1}{8}$	936	108	11.54	24.0	11.32
4	2 $\frac{1}{8}$	940	110	11.70	25.0	11.79
5	2 $\frac{1}{8}$	944	108	11.14	23.0	10.85
6	2 $\frac{1}{8}$	929	105	11.18	25.0	11.79
7	2 $\frac{1}{8}$	936	109	11.65	25.0	11.79
8	2 $\frac{1}{8}$	933	112	12.00	26.0	12.26
9	2 $\frac{1}{8}$	931	116	12.46	26.5	12.50
10	3	940	109	11.60	24.5	11.56
11	3	932	113	12.12	25.0	11.79
12	3	942	121	12.84	25.5	12.02
13	3	941	121	12.85	27.0	12.74
14	3	942	121	12.85	26.0	12.26
15	3	930	120	12.90	26.0	12.26
16	3	937	125	13.34	25.5	12.03
17	3	941	130	13.82	26.0	12.26
18	3	939	132	14.06	27.5	12.98
19	3 $\frac{1}{8}$	938	111	11.83	25.5	12.03
20	3 $\frac{1}{8}$	946	120	12.68	26.0	12.26
21	3 $\frac{1}{8}$	941	120	12.75	25.5	12.03
22	3 $\frac{1}{8}$	943	122	12.94	27.0	12.74
23	3 $\frac{1}{8}$	933	122	13.08	27.0	12.74
24	3 $\frac{1}{8}$	946	121	12.79	27.0	12.74
25	3 $\frac{1}{8}$	932	121	12.98	26.0	12.26
26	3 $\frac{1}{8}$	937	122	13.02	26.0	12.26
27	3 $\frac{1}{8}$	940	135	14.36	28.5	13.45
28	3 $\frac{1}{8}$	938	125	13.33	27.0	12.74
29	3 $\frac{1}{8}$	939	129	13.74	29.0	13.68
Average.....				12.51		12.16

Difference,

0.35 per cent

from 12.46 to 14.06. Sample 18, which measured only 3 inches of cream, actually contained more cubic centimeters of cream than either of the bottles with cream layers measuring 3 $\frac{8}{16}$ inches.

On the other hand the cream layer on sample 10, which likewise measured 3 inches, contained only 109 cc. of cream, or less cream than sample 4 with a cream layer measuring only $2\frac{14}{16}$ inches.

The data here given is not enough to show the range of variation to be expected in measurements of the cream layer from the top of the bottle but enough is given to make it plain that a variation of at least $\frac{1}{4}$ inch is necessary before one can be fairly sure which bottle contains the more cream. It should also be remembered that these measurements were made of the bottles of a single company which was striving to keep a stock of uniform bottles. Where comparisons are made between bottles of different companies the measurements of the cream layer from the top of the bottle are liable to be even more misleading.

The results obtained by marking the bottles and determining the cubic centimeters of cream present, when reduced to per cent by volume, correspond fairly well with similar figures from the test tube determinations. It will be observed, however, that the results obtained from the bottles range more widely than those from the test tubes. It has already been pointed out that average variation in the measurement of the cubic centimeters of cream in a bottle amounts to 0.7 per cent while the ordinary variation in the test tube determinations is less than 1 mm. which is equivalent to 0.47 per cent.

A comparison of the results as given by the two methods for similar bottles shows that in almost all cases the percentage of cream as determined by the test tubes is less than that determined direct from the bottle. The average of all these determinations in table 1 is 0.35 per cent by volume less for the test tube determinations. This variation while real is not very significant because it is only one-half of the normal variation in determining the volume of cream in milk bottles by direct measurement.

SUMMARY

The volume of cream in the milk bottle is important and there are wide differences of opinion as to the factors influencing this volume. This conflict of opinion is largely unsupported by

direct evidence and is primarily due to the lack of a method of measurement which is both accurate and readily applicable under milk plant conditions.

The improved method here described consists in filling round-bottomed test tubes, 1 inch in diameter, to a depth of 204 mm., 8 inches, with the milk to be tested. These tubes of milk are immediately cooled in ice water and when cool are held at 40°F. for approximately twenty hours. The depth of the resulting cream layer is measured in millimeters and each millimeter of cream represents 0.5 per cent of cream by volume. The volume of cream as determined in this way agrees closely with the volume of cream developed in milk bottles under similar temperature conditions.

This method has been extensively tested in milk plants and its advantage lies in the fact that by its use a large number of samples may be collected during a single day, the samples stored compactly, and measurements of the cream made quickly, accurately, and quantitatively.

ACKNOWLEDGMENT

In developing this method and testing its applicability to milk plant problems extensive observations have been made in the milk plants of the Gridley Dairy Company, of Milwaukee, Wis., the Sheffield Farms Company, Inc., of New York City, the Detroit Creamery Company, of Detroit, Mich., the Pevely Dairy Company, of St. Louis, Mo. and the Bowman Dairy Company, of Chicago, Ill. The opportunity thus afforded of testing the method under commercial conditions has added much to its value and acknowledgment is gladly given of the many courtesies extended. While in connection with each of these milk companies there have been a number who by their criticism and suggestions have aided in this and the accompanying studies mention is especially due Messrs. S. M. Heulings, of the Sheffield Farms Company, Inc., and W. D. Dotterer, of the Bowman Dairy Company.

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A WEIGHT-HEIGHT-AGE CURVE AS A MEASURE OF THE STATE OF NUTRITION AND OF GROWTH OF THE DAIRY COW

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The weight of an individual cow at a given age is, by itself, not an index of her condition or state of nutrition. The cow may naturally, by heredity, have a skeleton somewhat below or above the average in size and, therefore, because of the tendency to keep the symmetry between weight and height constant, also weigh less than the average. It is in fact very rare that an individual cow, selected at random, has the weight of an average cow. A ten-month-old Holstein heifer selected at random from our herd was found to differ by 50 pounds from the average weight of our ten-month-old Holstein heifers; and the difference in weight between the heaviest and lightest heifers of this breed and age in our herd was found to be 270 pounds. In other words comparing an average individual heifer under experimental conditions to the average weight of a heifer of the same age as a standard may introduce an average experimental error of 50 pounds due to the individuality of the animal and this may be as high as $\frac{270}{2}$ or 135 pounds, or even higher.

These illustrations indicate the inadequacy of using the average weight by itself as a standard of comparison for the determination of the condition of a cow under experimental conditions and suggests the necessity of adopting some standard of measure which will take into consideration what might be termed as the "hereditary size" of the animal. The ideal hereditary size of each individual, if known, would be the ideal standard of comparison for that individual under the given experimental conditions, in order to determine what effect such experimental conditions have on the animal.

Investigations at this station by Waters, Mumford, Trowbridge and their associates have shown that experimental conditions exert little or no effect on the growth in height at withers. This is strikingly illustrated by figures 1 and 2¹ showing the relative effect of experimental conditions on height and weight on the same group of animals. The effect on weight is profound; the effect on height at withers is practically negligible.²

If height at withers is practically independent of experimental conditions it may be taken as an unvarying measure of the hereditary size of the animal at any age. If the relation between height and weight under "normal" conditions is established, it is easy to determine from this established relation, knowing the height, what should be the corresponding weight. This is easiest accomplished by a weight-height curve as shown in figure 3.³ For every height there is under a given set of conditions, a corresponding definite weight. Knowing the height of the animal under experimental conditions the weight corresponding to this height is found from this curve for "normal" animals and compared to the actual weight. The difference between the two is a measure of the effect of the experimental conditions on the animal.

That there is a definite weight for a given height under "normal" conditions regardless of the hereditary growth capacity, is shown in figure 3. The height and weight of the relatively small Jersey fall on the same line as the weight and height of the much larger Holstein, and this is probably true for all dairy breeds. The relation of symmetry probably holds for all breeds of the same type. The only difference between the different breeds of different hereditary sizes, consists in the dif-

¹ The writers are indebted to Dr. C. R. Moulton, Chairman of the Department of Agricultural Chemistry in this station for figures 1 and 2. Further details concerning these animals may be found in Research Bulletin 43 of this station.

² In this connection it is important to note that investigations at this station by Eckles, reported in Research Bulletins 31 and 36, have shown that from birth to maturity the dairy animal increases only 90 per cent in height at withers while in the same period of time it increases, approximately, 1423 per cent in weight.

³ Figure 3 is based on data in Research Bulletin 36 of this station by C. H. Eckles.

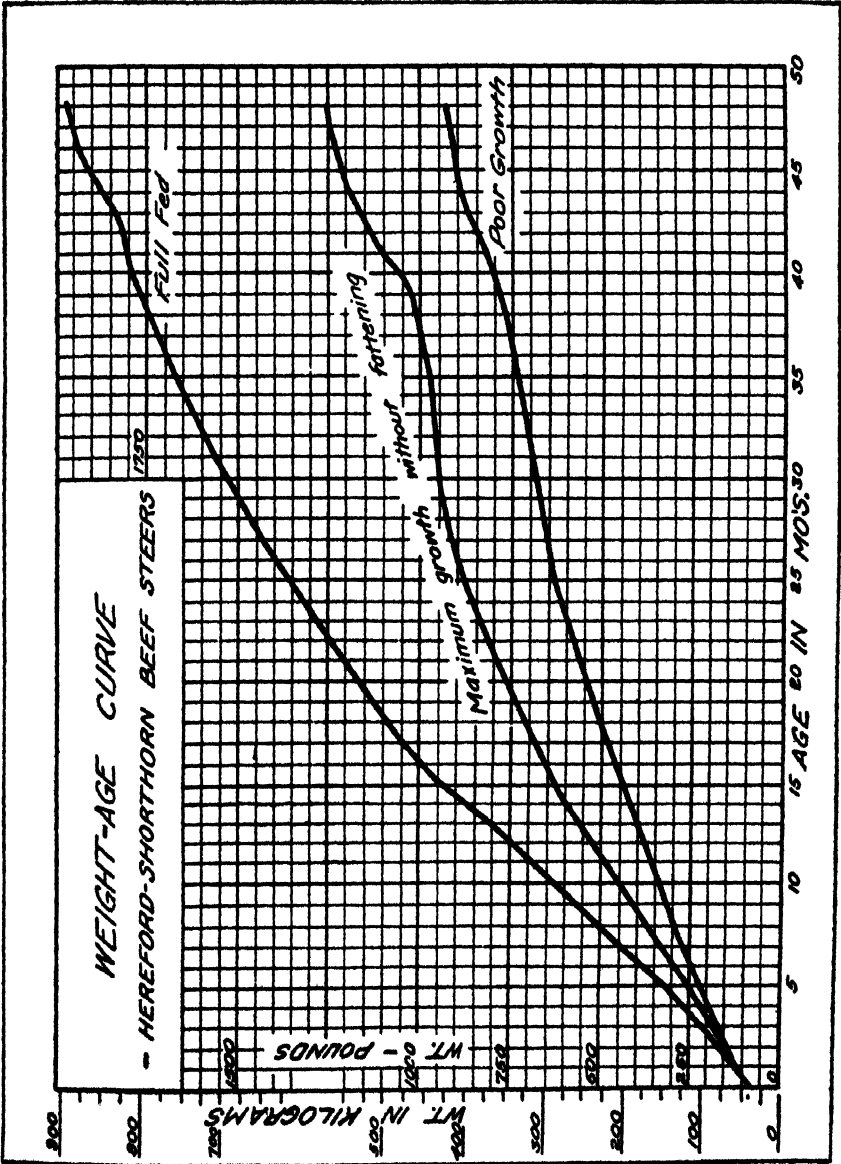


Fig. 1

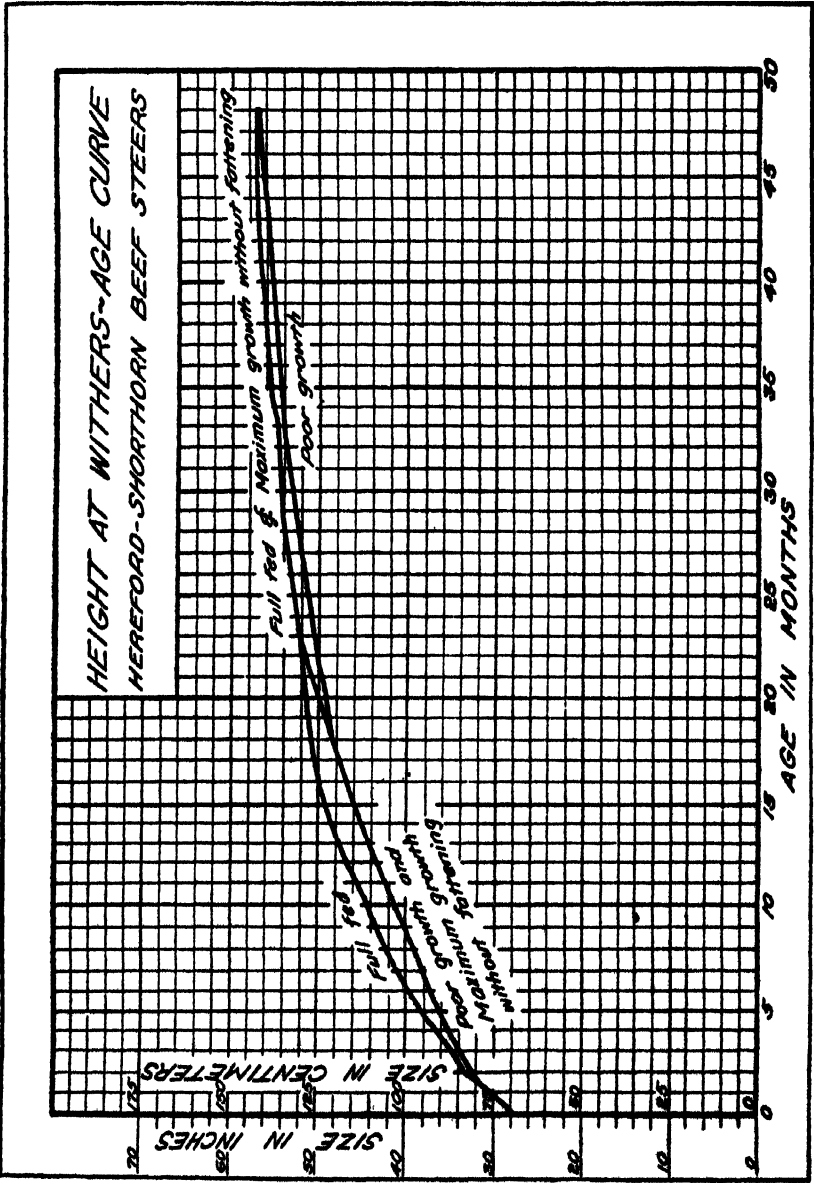


FIG. 2

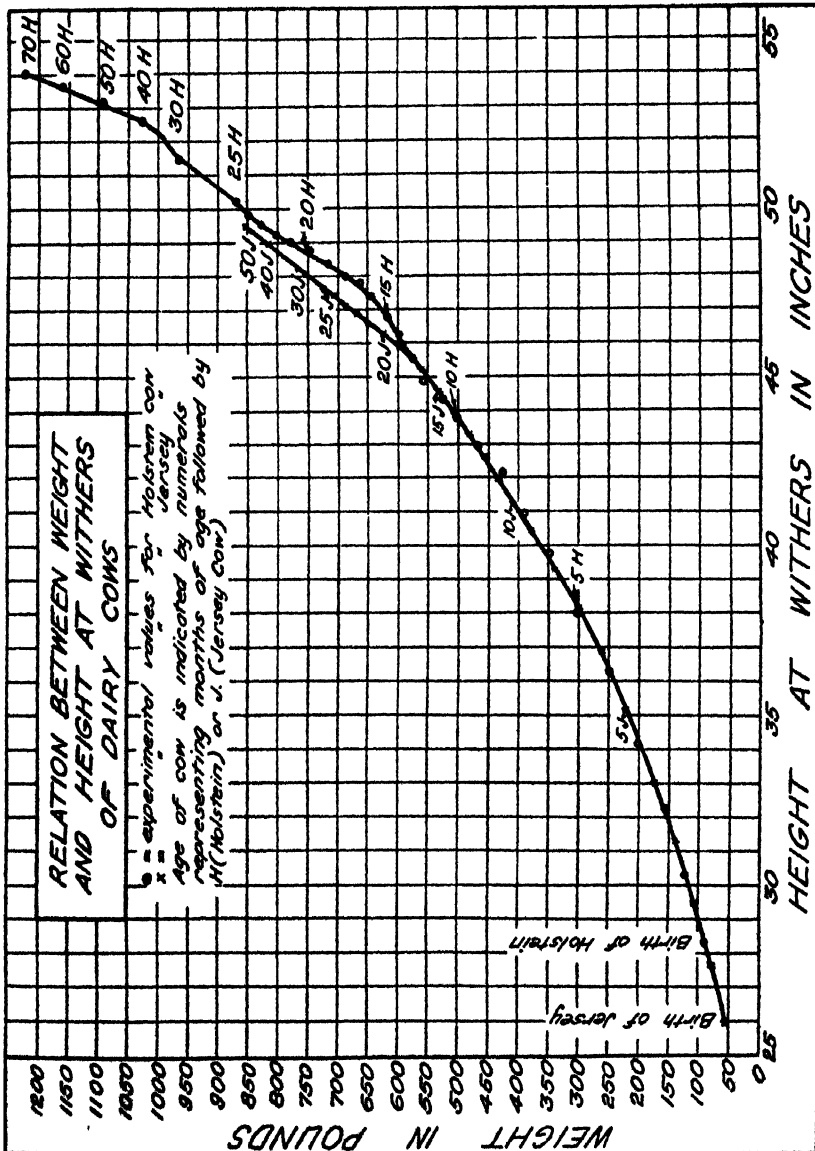


FIG. 3

ference between the age-height relationship and age. Thus from figure 3, a fifteen-month-old Jersey has the weight-height relation of a ten-month-old Holstein; a fifty-month-old Jersey has the weight-height relation of a twenty-four-month old Holstein. But for a given height, for example, 45 inches, there is always the same corresponding weight of 550 pounds, regardless of the breed.

This curve, therefore, eliminates errors due to individuality of the animal, taking the "ideal hereditary" size of the given animal as the standard unit for its measure and serves not only as a measure of the state of nutrition of the animal but also as a measure of its growth as compared to the average animal of the given breed or race. Incidentally the curve may also be used for determining the weight or height at any age for the Holstein or Jersey cow.

ACTINOMYCES IN MILK WITH SPECIAL REFERENCE TO THE PRODUCTION OF UNDESIRABLE ODORS AND FLAVORS¹

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To the knowledge of the author it has not been heretofore suggested that actinomyces may cause certain obnoxious tastes and odors in dairy products. The mere presence of these organisms in milk and other foods has scarcely been noted, while their activities are still less known. Only a few references of direct connection to this paper are considered here.

Gratz and Vas (7) described two species of casein-dissolving actinomyces from cheese. Coombs (4) found the number of fungus spores in normal and moldy butter greatly decreased in number during ageing. It is probable actinomyces as well as molds play a part in causing abnormal flavors in butter for the odors and tastes produced by actinomyces are if anything more penetrating than those of molds. Thom (14) has shown that fungus spores find their way into milk mainly from fodder and dust. Some spores pass through the separator and thus gain access to the butter. Conn (3) described several actinomyces-like organisms such as *B. arborescens lactis* n. sp., *Bacillus* 138 and *Bacillus* 184. These organisms imparted to butter made from inoculated milk, undesirable flavors. Moore (10), Dykstra (6), and others have stated the natural habitat of *A. bovis*, the ray fungus is in the soil, and that the method of entrance to the body is by a cut or abrasion about the head. This organism is not known to be transmitted to the milk. Waksman

¹ Work done at N. J. Agr. Exp. Stas., New Brunswick, N. J. Acknowledgment is due Dr. J. G. Lipman, Director, and Mr. L. S. Riford formerly Assistant Dairy Husbandman for criticism and encouragement during the progress of the investigation.

(16) has established the identity of morphology, cultural characters and biochemical reactions of the so-called saprophytic and parasitic species. Practically the only differences lie in the temperature relations and pathogenicity. He has also shown that while many species grow well in milk as a medium, only some of them cause coagulation and peptonization. Some species which he studied were able to utilize both the carbon and nitrogen of the casein. Conn (2), Krainsky (9), Munter (11), Rossi-Doria (12) and Waksman and Curtis have contributed much of value to the literature regarding this group of organisms.

EXPERIMENTAL

This investigation deals with an outbreak of an offensive taste and aroma developing in milk at a New Jersey dairy. From sixteen to forty-eight hours was necessary for the repulsive taste to develop. At first it resembled somewhat strong turnipy milk later developing the characteristic earthy and bitter-moldy taste and odor. The dairy was well managed and equipped to produce milk and cream of excellent quality. Utensils were sterilized twice daily, the cows were cleaned daily, the bedding was baled straw while the feed consisted of alfalfa, sedge grass and grain. The fodder was grown on low mucky land and had a somewhat earthy odor.

Samples of the milk and cream developing the offensive taste were plated bacteriologically on lactose litmus agar and incubated at 25°C. for several days. The plates were found to be populated largely by actinomyces colonies, the average of the six samples examined being 1,100,000. A sample of cream gave a count of 2,400,000 per cubic centimeter. In these samples no differentiation was made between actinomyces and bacterial colonies. Another sample of cream plated on Czapek's fungi agar (5) and incubated for eight days at 25°C. yielded 1,100,000 actinomyces and 1,000,000 bacteria per cubic centimeter. In still another series of samples the counts were 440,000 actinomyces and 120,000 bacteria. The offensive taste was shown to be due to a biological factor because it required from sixteen to forty-eight hours for the taste to develop after inoculating

sterile or normal fresh milk with very small quantities of the abnormal milk. Such inoculated samples invariably revealed the presence of large numbers of actinomyces after plating on Czapek's agar. This medium gave excellent satisfaction and on it actinomyces develop aereal mycelium and otherwise appear more characteristic than on ordinary peptone-beef with or without sugars. It is believed that the lack of suitable media, the short incubation period usually employed, and the confusion of young actinomyces colonies with bacteria, are probable reasons why this group of organisms in milk and in other food substances, has been largely overlooked.

The trouble gradually subsided during the summer when the cows were receiving fresh feed, but recurred again in the fall. Again the dairy was inspected, samples taken, and the cause of the bitter-moldy taste traced to the large number of actinomyces present. Samples packed in ice and examined bacteriologically after twenty-four hours, showed, 1,000,000 actinomyces and 450,000 bacteria per cubic centimeter for milk, and 1,800,000 actinomyces and 700,000 bacteria per cubic centimeter of cream. Samples of milk from six cows in the dairy were drawn directly into sterile flasks under regular milking conditions, packed in ice and examined after about twenty-four hours. The cows were brushed with a cloth and given clean straw for bedding a short time before milking. Samples of the hay, grain, straw, manure, water and litter, were also taken to the laboratory for bacteriological examination.

Table 1 contains data regarding the milk drawn from three cows of the herd under investigation as well as several other representative herd and market samples for comparison. Czapek's agar with a seven-day incubation at 25°C. was used in this work. The average per cent of actinomyces in the normal milk examined was about 2.5; the minimum, 0, and the maximum 10 per cent. Practically all raw milk contains some of these organisms because of the practical difficulties preventing dust, soil and bedding contamination of the milk. From table 1 it is evident that the milk of three cows in the affected herd contained a sufficient number of actinomyces to give a distinct

TABLE 1
Relative numbers of actinomycetes and bacteria in milk

SOURCE OF SAMPLE	TASTE FRESH	TASTE TWENTY-FOUR HOURS	TASTE FORTY-EIGHT HOURS	BACTERIA PER CUBIC CENTIMETER, CONN'S ACTINOMYCETES AGAR, FORTY EIGHT HOURS, 25°C.	ACTINOMYCETES PER CUBIC CENTIMETER, CONN'S ACTINOMYCETES AGAR, FORTY EIGHT HOURS, 25°C.	CHARACTER OF SAMPLE IN FOUR DAYS	AMOUNT REQUIRED TO PRODUCE TASTE IN 100 CC. MILK IN TWENTY-FOUR HOURS AT 25°C.
Primrose 3rd (affected herd)	Normal	Bitter-earthy	Bitter-moldy, marked	450,000	700,000	Curdling just beginning; intense bitter-pungent taste	cc. 1
Florham Fashion (affected herd)	Normal	Oily-earthy	Bitter-oily	50,000	210,000	Curd precipitated; whey clear; slight brown color, very bitter	1
King's Pride (affected herd)	Normal	Normal	Flat-earthy	260,000	650,000	Solid curd, normal souring	10
Mixed milk College Farm herd	Normal	Normal	Slightly sour	500,000	8,000	Normal souring	0
College Farm herd	Normal	Normal	Slightly sour	220,000	3,000	Gassy curd	0
College Farm herd (after sweeping)	Cow	Normal		200,000	220,000	Gassy curd partly digested	0
Creamery New Brunswick, N. J.	Normal	Normal	Normal souring			Normal souring	0

Dairy at New Brunswick, N. J.	Normal	Slightly sour	Sour	400, 000	8, 000	Solid curd, some gas	0
Dairy at Charlotte, N. C.	Normal	Normal	Normal	60, 000	500	Normal curd	0
Dairy at Charlotte, N. C.	Normal	Normal	Normal	120, 000	2, 000	Normal curd	0
Dairy at Columbus, Ga.	Normal	Normal	Sour	280, 000	6, 000	Slightly gassy curd	0
Dairy at Seattle, Wash.	Normal	Normal	Slightly sour	40, 000	3, 000	Firm curd	0

earthy flavor to the milk held at air temperature for twenty-four hours. In thirty-six hours the milk was so offensive to the taste that it would have been impossible to use it. The taste is described as bitter-moldy, but the term "actinomyces taste" is suggested as being more descriptive and correct. In two of the three samples the curd was precipitated in four days with partial digestion; in the third sample no curdling occurred. Of the first two samples above, only 1 cc. was necessary to impart to normal sterile milk an actinomyces taste in twenty-four hours. Counts made by the author on various market milks, show the numbers of actinomyces present is relatively low. The affected herd's water supply contained only 2 actinomyces per cubic centimeter.

Table 2 contains data relative to the numbers of actinomyces in various materials. In order to determine the source of the actinomyces, straw, hay, grain, soil, muck, dust, and manure were examined bacteriologically. This data is found in table 2.

From table 2 it is evident that actinomyces are widely distributed in nature, particularly in dry hay, straw, dust and soil. Muck soils and sod contain more of these organisms than dry fallow or cultivated soils. The spore-like bodies of actinomyces are very light and remain in the air for a long time. This was proven both in the laboratory and in the stable. Petri dishes exposed for thirty minutes after raising dust contained many actinomyces colonies—the proportion of the latter to bacteria being greater after partial subsidence of the dust. Neither ground nor surface waters appear to carry many actinomyces. The hay and straw from the dairy producing the bitter-moldy milk, contained an unusually large number of these organisms and it is extremely likely that dust contamination in the stable before or at the time of milking was responsible for the trouble.

Over 30 types embracing 11 species were isolated and identified from the milk. These same species and many additional ones were identified from soil, hay, straw and dust. Some of the organisms were very slow growing, some produced no aerial mycelium and grew only upon special media and still others produced no characteristic changes in milk. Waksman (15,

TABLE 2
Numbers of actinomyces in various substances

SUBSTANCE	MEDIUM USED	ACTINOMYCES PER GRAM
Ground alfalfa hay	Conn's actinomyces agar incubated seven days, 25°C.	3,900,000
Timothy and clover hay	Conn's actinomyces agar incubated seven days, 25°C.	2,300,000
Redtop and timothy hay	Conn's actinomyces agar incubated seven days, 25°C.	600,000
Chopped wheat straw	Conn's actinomyces agar incubated seven days, 25°C.	390,000
Shredded oat straw	Conn's actinomyces agar incubated seven days, 25°C.	90,000
Dried manure	Conn's actinomyces agar incubated seven days, 25°C.	145,000
Wet manure	Conn's actinomyces agar incubated seven days, 25°C.	1,500
Ground oats	Conn's actinomyces agar incubated seven days, 25°C.	190,000
Ground mixed feed	Conn's actinomyces agar incubated seven days, 25°C.	6,000
Dried soy beans	Conn's actinomyces agar incubated seven days, 25°C.	8,000
Single soy bean seed having musty odor	Ashby's nitrogen-free agar	95,000
Five alfalfa seeds	Ashby's nitrogen-free agar	3,600
Soil sample 1	Conn's actinomyces agar	100,000
Soil sample 2	Conn's actinomyces agar	70,000
Soil sample 3	Conn's actinomyces agar	150,000
Muck soil	Ashby's nitrogen-free agar	350,000
Muck, Farmogerm legume inoculant	Ashby's nitrogen-free agar	1,800,000
Muck soil, Alphano, N. J.	Ashby's nitrogen-free agar	750,000
Grass roots from muck soil	Conn's actinomyces agar	475,000
Average of four water samples	Conn's actinomyces agar	3
Dusty air, five minutes exposure	Conn's actinomyces agar	20,000 (10 per cent of total colonies)
Dusty air, ten minutes exposure in stable	Conn's actinomyces agar	76,000 (16 per cent of total colonies)

16, 17) has published complete descriptions of 41 species. He noted their growth on milk, action on casein and enzyme production. In this particular study the most frequently encountered organisms were closely similar types of the *A. griseus* group, represented by *A. griseus* Krainsky. Because of its importance a somewhat complete description of this species is reproduced here. Krainsky's original description is very incomplete.

Actinomyces griseus Krainsky

Czapek's agar. Colonies round, raised, 2 mm. diameter in four days, color water green (G. tint 2).² Reverse, sage yellow to lemon yellow (G. Y. tint 2 to O. Y. normal tone) in old cultures. White aerial mycelium, formed early changing to water green color. Powdery colonies. Old cultures become a light fluorescent green color.

Microscopically the colony appears as fine radiating many branched filaments. The latter break up into short chains or rods of conidia each conidial chain being from 1 to 2.2 micra long and from 0.6 to 0.8 micron broad. On staining these show oval spore-like bodies 0.8 to 1.2 micra long by 0.5 to 0.9 micron broad, which are highly refractive and stain with difficulty. They occur in chains, pairs or branching chains preserving the form of the original filaments. Odor of culture, strong actinomyces (musty straw).

Bouillon agar. Abundant but uncharacteristic growth in three days. Colonies round, much raised and white. Aerial appears late, chalky white at first becoming darker with age. Enzymic zone or halo extends around the colony. Medium only very slightly darkened. Reverse color brownish (Y. O. shade 2); odor, moderately strong actinomyces.

Conn's glycerin actinomyces agar (2). Colony diameter in three days about 2.5 mm. and in six days from 3 to 5 mm. Color, white at first, later becoming water green with a gray mycelium in the substratum. Reverse (G. Y. tint 2); old colonies much zonated. Few or no spiral structures in aerial mycelium. Microscopically like *Czapek's agar*. Hyphae stain well with methylene blue, the conidia appearing in chains within the filament. Growth on this medium is poor and not characteristic when either the glycerin is omitted or the reaction is made neutral.

² See color charts accompanying Mulliken's "Identification of pure organic compounds."

Krainsky's calcium malate agar (9). Growth similar though not as vigorous as on Conn's medium, scanty at first then spreading. Aerial mycelium white to gray at first later becoming pale green (Y. tint 2); reverse (O. tint 2).

Starch agar. Growth poor even after ten days; diastatic activity weak.

Gelatin (15 grams in 1000 cc. distilled water). Colony diameter in four days, 1 mm. while in seven days it is 1 to 1.5 mm. with a liquefied pit from 4 to 6 mm. diameter. The growth is grayish and the liquefied portion shows a slight yellow-green fluorescence, becoming darker with age. Aerial mycelium slow to form, scanty, white, with no spiral bodies. Growth more vigorous on acid than on neutral gelatin. Pigment is deeper on neutral media. Color, yellow to golden yellow (Y. to golden Y.).

Potato. Vigorous growth; brownish white, abundant aerial mycelium becoming greenish colored about the seventh day. Odor, moderately strong actinomyces. Potato darkened slightly in three days, the color increasing in intensity with age.

Czapek's solution. In four days the medium assumes a green color and is filled with white flakes. Colonies appear on the surface in from seven to twelve days, they also appear on the bottom and sides of the flask. Odor, moderately strong actinomyces.

Milk. A bitter moldy taste and earthy odor develops in from twenty-four to forty-eight hours. No growth evident macroscopically for about three days, when colonies develop on sides and bottom of flask. Surface colonies appear in from six to ten days, the color is white at first and gray later. In from three to five days the odor becomes very strong and the taste extremely bitter-moldy; the casein is coagulated in from three to seven days, active peptonization then begins and is practically complete in from ten to twelve days. The reaction becomes alkaline.

Nitrate reduction. The reduction of nitrates to nitrites in a peptone-nitrate medium was relatively weak. The transformation of protein into ammonia however was strong.

Indol production. Negative.

Nitrogen transformation. In four days at 25°C., this organism ammonified 3.04 per cent of the total nitrogen contained in whole milk. Under similar conditions, 4.88 per cent of the total nitrogen contained in a 1 per cent casein solution was converted into ammonia. The initial acidity in terms of lactic acid was 0.18 and after four days the reaction was alkaline.

Besides *A. griseus*, the following species were identified from milk; *A. albus* Krainsky, *A. alboflavus* Waksman and Curtis, *A. flavus* Kriaisky, *A. lipmani* W. and C., *A. albosporeus* Krainsky, *A. diastaticus* Krainsky, *A. rutgersensis* W. and C., *A. bobili* W. and C., an organism corresponding in some respects to *A. violaceus* W. and C. and a slow growing species which formed no aerial mycelium on any media. This organism was very active in milk and was apparently a new species. All of these organisms produce a bitter-moldy taste in milk but the more active species are *A. griseus*, *A. albus*, *A. rurgersensis* and *A. lipmani*. From twenty-four to forty-eight hours is usually required for the odor to develop, although the taste is evident in a shorter time. It is possible therefore for milk heavily contaminated with actinomyces, to become offensive before it reaches the consumer. Refrigeration materially delays but does not absolutely inhibit the formation of the objectionable taste. The character of the bitter substance is unknown; it is probably due to extracellular enzymes or other soluble bodies elaborated by the organisms during active growth. Usually there is no macroscopic change in milk for several days then more or less complete coagulation of the casein followed by peptonization.

ACTINOMYCES IN OTHER FOODS

To the author's knowledge actinomyces have not been mentioned as a cause of stale, musty or moldy odors and flavors in foods or food materials. It is certain many of the odors and tastes commonly attributed to molds are really due to actinomyces. The author has twice examined musty smelling walnut meats and found the cause to be actinomyces. Both spores (conidia) and mycelium were present and the species resembled closely *A. griseus*, though not entirely identical with it. In another case, dried fish flakes were found to be stale and musty smelling. Bacteriological examination revealed the presence of both actinomyces and molds. Upon reinoculation however only those flakes inoculated with actinomyces reproduced the

characteristic odor and taste. The mold was therefore deemed of secondary importance, though its penetrating powers were greater than the actinomyces.

Oats, barley, corn and ground cereals often become moldy, particularly when excessive moisture is present. Several species of aspergillus, penicillium and fusarium are often present under such conditions. Thom (13) has reported the presence of many species of molds and actinomyces in normal and spoiled corn meal. The author has examined several samples of oats and barley which contained large numbers of actinomyces, molds, spore-bearing bacteria and *B. aerogenes*. It is possible actinomyces may play a part in the development of musty odors in grains under certain conditions. Dried eggs often contain many actinomyces and it has been suggested³ that the moldy odor and taste often encountered in this product may be due to this group of organisms. They thrive well in dry places, are not readily killed by desiccation and may utilize a wide variety of materials for their food. The odors and tastes produced by them are intense and objectionable and easily permeate the substratum in which they are growing, and for these reasons are likely to cause trouble in a variety of products.

SUMMARY

Actinomyces are often present in market milk samples. In normal samples these organisms constituted about 2.5 per cent of the total organisms present. In abnormal samples, particularly those drawn from cows in dusty stables, this figure may reach 50 per cent.

The principal means of entrance to milk are hay, straw, grain, soil and dust raised from these materials.

Actinomyces may under certain conditions cause an obnoxious bitter-moldy taste to develop in milk after some hours storage. The term "actinomyces taste or odor" is suggested to describe the characteristic taste and odor of actinomyces.

³ Personal communication, Dr. H. W. Redfield, Chief, N. Y. Station, U. S. Bureau of Chemistry.

Two outbreaks of bitter-moldy milk in the same dairy were traced to the presence of large numbers of actinomycetes. The source of the contamination was probably hay and straw.

Eleven species were identified from the milk. The two most active species concerned were *A. griseus* group and *A. albus*. These organisms grow readily in milk and are able to produce profound changes in the casein and whey. The extremely diffusible and volatile substance which causes the odor and taste so characteristic of actinomycetes is not known.

Actinomycetes may cause stale, musty and moldy odors in such foods as walnuts, dried fish, cereal grains and possibly dried eggs. They occur together with molds in many foodstuffs, especially those in a dried condition.

In studying actinomycetes or in attempting to isolate them from foods or other materials, synthetic media combined with long low temperature incubation should be used. The ordinary methods and media with incubation at 37°C., are unsatisfactory.

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PLATE 1

FIG. 1. *A. rutgersensis* Waksman and Curtis; old culture on Czapek's agar.

FIG. 2. *A.* no. 2, an undescribed species; compact, colorless colonies, scant aerial mycelium.

FIG. 3. *A. griseus* Krainsky, old culture on Czapek's agar.

FIG. 4. *A.* no. 4, similar in some respects to *A. violaceus-caesari*, W. and C. Conn's agar, twenty days.

FIG. 5. *A. diastaticus* Krainsky, twenty-day culture on Czapek's agar.

FIG. 6. *A. violaceus-ruber* Waksman and Curtis, old culture on Ashby's nitrogen free agar.

FIG. 7. Same as figure 6; twenty-day culture showing aerial mycelium on Czapek's agar.

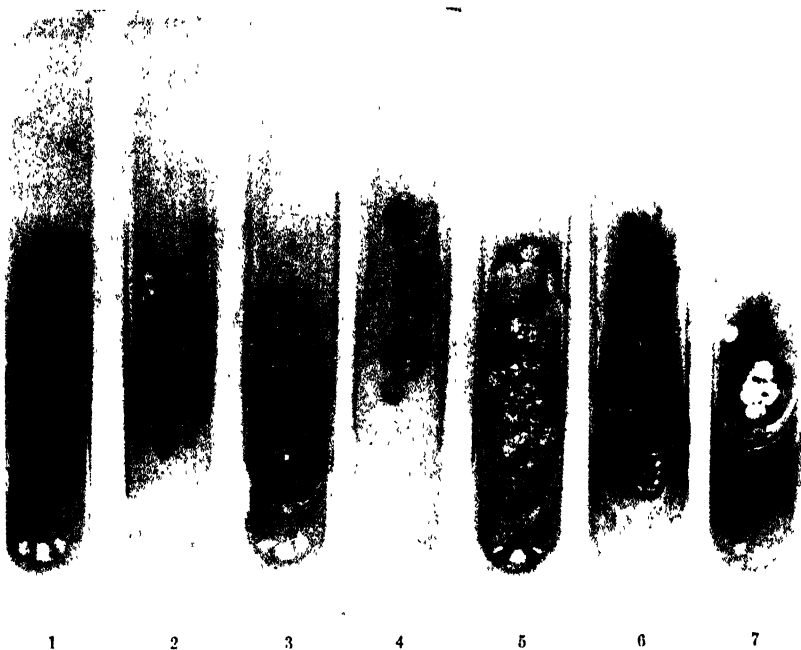


PLATE 2

FIG. 1. *A. flavus* Krainsky, old culture on Krainsky's calcium malate agar.

FIG. 2. *A. lipmani* Waksman and Curtis, showing abundant, spreading growth with gray aerial mycelium on Conn's agar.

FIG. 3. *A. albus* Krainsky, fifteen-day growth on Conn's agar showing dense aerial mycelium.

FIG. 4. *A. boboli* Waksman and Curtis, Conn's agar, coral-red colony with no aerial mycelium.

FIG. 5. A culture resembling somewhat *A. albosporeus* Krainsky, aerial mycelium formed late if at all. Growth on Conn's agar after thirty days.

FIG. 6. *A. albus* Krainsky, Czapek's agar after fifteen days showing prolific aerial mycelium.



THE CARE OF MORNING MILK BEFORE PASTEURIZATION¹

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The quality of the raw milk supply is always a matter of concern to the operators of pasteurizing plants. Fortunately those located in the smaller cities are in a much better position to improve it than are those in the larger cities because they are nearer to the source of supply. One of the problems, which is often confronted, is whether or not it is necessary for the farmer to cool his morning's milk before it is brought to the plant for pasteurization. In most of these smaller cities the farmer is located within a few miles of the plant and makes his own deliveries within a few hours after the morning milking. During the summer season he is always in a hurry and if the time and labor involved in cooling the milk could be eliminated he would feel relieved. The actual necessity for cooling this morning milk is often questioned by the producer. The dealer and the health officer are themselves sometimes undecided about it. The frequency with which the advice of the dairy division has been asked on this point has led to the tests here reported.

PURPOSE OF INVESTIGATION

With this problem in mind, a brief study was made to determine the practicability of such a plan. The study did not contemplate a further investigation of the so-called "germicidal" property of fresh milk, nor was any elaborate preparation of utensils or control of other conditions attempted. The purpose of the work was to note what changes might take place in the bacterial count of an average can of milk as it might be found on any farm at the time of milking or during the period

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previous to delivery at the plant for pasteurization. No effort was made to select any particular milk or cans. They might have represented those from any average plant or farm, no better and no worse. Some cans were clean and dry, others fairly clean but damp. On some days the temperature of the outdoor air was high, on others moderate. The conditions represented ordinary daily variations during the summer months.

METHOD OF PROCEDURE

At the time of the morning milking, a can was chosen at random from those ready for use in the milk room. It was filled at once with fresh milk. A sample of this was plated immediately on standard lactose agar. The plates were incubated at 37° for forty-eight hours. The temperature of the milk was noted and recorded.

The covered can was allowed to remain undisturbed at the temperature of the room during the remainder of the period. Subsequent platings were made at intervals of one hour for five consecutive hours, and the temperatures recorded.

EXPERIMENTAL DATA

The bacterial counts of the samples, arranged according to the initial counts, are given in table 1.

It will be noted that there is a marked variation in the bacterial content of the fresh milk, a few representing milk of excellent quality, while the majority show a remarkably high count. It may be said that the majority of the cans were damp, as might be the case on the average farm. A number of the samples show a decrease in the count during the first two or three hours. They are distributed widely. Of the 24 samples, at the end of one hour, 10 had a lower count than the original; after two hours, 8; and after three hours, only 1.

A bacterial count of 1,000,000 per cubic centimeter by the agar plate method, is accepted in many localities as the maximum for milk to be used for pasteurization.

In looking over the data, it may be noted that 50 per cent of the samples exceed this standard at the end of four hours. Those

TABLE 1
Bacterial count by agar plate method

SAMPLE NUMBER	BACTERIAL COUNT PER CUBIC CENTIMETER					
	Original	After one hour	After two hours	After three hours	After four hours	After five hours
1	1,770	1,780	2,000	2,070	3,100	8,370
2	1,820	880	1,340	3,900	6,700	8,000
3	6,800	1,800	4,400	3,100	12,500	19,600
4	10,900	8,900	2,200	29,600	33,900	42,000
5	12,500	18,800	24,000	35,000	25,000	50,000
6	14,900	17,200	19,300	53,000	88,000	201,000
7	104,000	214,000	290,000	296,000	410,000	900,000
8	109,000	83,000	81,000	191,000	470,000	1,600,000
9	138,000	146,000	102,000	246,000	288,000	570,000
10	161,000	143,000	353,000	375,000	1,600,000	2,340,000
11	176,000	180,000	244,000	325,000	370,000	530,000
12	177,000	320,000	490,000	1,000,000	1,980,000	5,500,000
13	181,000	332,000	380,000	410,000	1,400,000	2,250,000
14	205,000	253,000	298,000	330,000	600,000	770,000
15	219,000	200,000	215,000	480,000	1,760,000	2,860,000
16	224,000	215,000	270,000	800,000	1,590,000	2,800,000
17	241,000	320,000	470,000	1,000,000	2,600,000	4,400,000
18	243,000	275,000	400,000	1,120,000	1,600,000	2,460,000
19	287,000	370,000	430,000	1,510,000	3,300,000	5,500,000
20	370,000	470,000	600,000	1,550,000	4,500,000	10,200,000
21	422,000	230,000	97,000	730,000	780,000	740,000
22	480,000	330,000	270,000	1,670,000	2,880,000	4,500,000
23	760,000	910,000	1,400,000	4,900,000	6,700,000	9,500,000
24	1,390,000	540,000	2,320,000	3,800,000	3,700,000	9,200,000
Average	247,320	232,515	365,135	869,153	1,529,050	2,789,540

with an original count of less than 100,000 per cubic centimeter are still well below this limit even after five hours.

The 24 samples may be divided into groups of 12 on the basis of the original count as follows:

	AVERAGE COUNT	
	Original	Final
Lowest (12).....	76,140	418,500
Highest (12).....	418,500	4,593,166

From this it will be noted that the group showing the lower original count also shows the lower final count.

In a comparison of an average of the 12 samples showing the lowest final count, it will be found that they also show the lower average original count.

Table 2 is a summary of the temperatures observed at the various intervals. The decrease in temperature is most marked during the first hour and very gradual during the remainder of the period, changing but slightly after the third hour.

TABLE 2

Range of temperatures at each interval during the five-hour period

PERIOD	MAXIMUM	MINIMUM	AVERAGE
	°F.	°F.	°F.
Original.....	96.8	86.0	92.1
After one hour.....	93.2	72.5	84.5
After two hours.....	87.8	72.5	82.0
After three hours.....	86.0	73.4	80.6
After four hours.....	85.1	72.5	80.0
After five hours.....	86.0	72.5	79.7

The relation between the decrease in temperature during the five-hour period and the original and final counts is shown in the following:

NUMBER OF SAMPLES	AVERAGE DECREASE IN TEMPERATURE	AVERAGE COUNT	
		Original	Final
	°F.		
12	10.5	143,395	1,663,000
12	14.1	96,381	1,126,544

The temperature throughout the period is such that it would be favorable for very active growth of most species. Consequently the slight difference between the two groups from the standpoint of the decrease in temperature is relatively insignificant. The initial count of the sample appears to be of much greater importance than the temperature.

SUMMARY AND CONCLUSIONS

The results of the study would indicate, as already generally understood, that the change in the bacterial content of milk during the first few hours after milking will depend upon:

1. The so-called "germicidal property" of certain individual samples.

2. *The initial count* (contamination from all sources).

3. The temperature at which it is held.

4. The time elapsing.

If milk is obtained from reasonably clean cows which do not give a high initial count (i.e., normally or because of any inflammation of the udder such as mastitis), milked into clean, *dry* utensils (especially milking machines and cans) and delivered to the pasteurizing plant within five or six hours after milking without cooling, the bacterial content should not be excessive for use for pasteurization. However, under average farm and plant practices, these conditions are not always to be found, particularly the clean, dry utensils. If the utensils are not clean and dry the initial count will tend to be so high that it will be impossible for the dairyman to deliver a milk which is satisfactory for pasteurization without cooling it adequately at once. In communities where the conditions can be satisfactorily controlled the farmer might be permitted to deliver uncooled morning milk to the pasteurizing plant. However, as a general practice in all communities it is not to be recommended.

LOSS IN THE GRAIN OF CORN IN STORAGE AS SILAGE

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The principle of ensiling corn involves two possibilities of loss: (1) Utilization of the silage may necessitate the feeding of more grain than is profitable; (2) the grain itself may suffer a loss of nutrients.

Emphasis is placed on the grain since it constitutes two-thirds the nutritive value of the crop and practically its entire merchantable value. (Reference is made to the present extensive practise of growing and harvesting corn for silage in such way as to secure a maximum yield of sound grain.) Nevertheless, corn is harvested as silage not as a better method of harvesting the grain or to enhance its feeding value, but purely to better utilize the stalk for feeding purposes. The continued and increasing use of corn silage is, of course, the best of evidence that the superior two-thirds of the crop represented by the grain is not seriously injured simply to make better use of the inferior one-third represented by the stalk.

At the time of harvest the grain has a definite cash value. Immediately upon harvest this value is destroyed since it is no longer merchantable. It still retains a feeding value but must be fed in such amount as is present in the silage ration. An ordinary silage ration of 35 pounds contains the equivalent of 5 pounds of dry shelled corn. It may be unprofitable to feed corn in such amount depending on the price of corn and the feeding use made of it. When corn was selling above a dollar a bushel the point became acute and a continuation of the high price level would have undoubtedly greatly curtailed the use of the crop for silage. So far as the dairyman is concerned he will usually want to feed corn in as large amount as that present in the silage ration and so is not particularly concerned with this feature of possible loss.

There remains the second possibility, namely, loss of nutritive value in storage. The present paper gives a limited amount of data bearing on this point but is offered more particularly as giving a method of attacking the problem. The method is based on the fact that in the mechanical preparation of the crop the kernels are practically all shelled from the cob and the majority pass into the silo intact. Some are cut and crushed but those which escape uninjured should be representative of the whole. By taking the kernel as a unit and using a number sufficiently large it is possible to determine the changes occurring in the grain under perfectly normal conditions of storage. The number of kernels to be used will be determined by the variability of the kernels themselves in respect to the qualities under study.

In a preliminary way, the dry matter changes in the grain as stored in silo and crib have been determined on the basis of the above method. A wood stave silo was filled September 28 and 29, 1920, from a field of Silvermine corn. Representative rows of the field were left standing and the corn husked and cribbed later. The yield of grain, estimated from these rows was 55 bushels per acre. The yield of silage was about 9 tons per acre. At the time of filling two samples of grain were collected from the silo and the positions marked by boards placed in the silage. When the silage was fed down to the board markers, June 25, and August 10, 1921, samples of grain were again collected. From the grain samples, lots of 100 kernels, intact but otherwise selected at random were used as subsamples. The cribbed corn from the rows left standing in the field was shelled March 16, 1921, and sampled. Lots of 300 kernels selected at random were taken as subsamples. Moisture was determined by the Brown-Duvel method.¹ Results are given in the table.

The table shows the kernel takes up moisture in storage as silage and that the percentage water content is increased from 35.85 as stored to 39.44 after storage. Evidently a physical

¹ Circular no. 72, Bureau of Plant Industry, U. S. Department of Agriculture. The method was modified by holding the temperature at 110°-115° for ten minutes with the high moisture samples in order to prevent excessive oil being carried over by the rapid evolution of steam.

balance is established with reference to water between the kernel and its surrounding medium. Moisture content of the silage was not determined but was probably around 70 per cent. Kernels saturated with water by soaking thirty hours contained 46.35 per cent of water.

TABLE 1

Loss of dry matter in the grain of corn as harvested and stored in crib and silo

	NUM- BER OF KER- NELS USED	AVERAGE PER KERNEL				LOSS OF DRY MATTER
		Weight	Dry matter	Water	Dry matter	
		<i>grams</i>	<i>grams</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
As harvested in silo.....	1900	0.4573	0.2934	35.85	64.15	
After storage in silo.....	1500	0.4599	0.2785	39.44	60.56	5.08
After storage in crib.....	1200	0.3382	0.2868	15.20	84.80	2.25

The per cent of dry matter in the grain is decreased by the absorption of water, but there is also an actual loss of dry matter. Based on the dry matter present at time of harvest this loss amounts to 5.08 per cent. The cribbed corn also showed a loss of dry matter amounting to 2.25 per cent on the same basis. The loss in the silo appears to be greater than in the crib but both are small.

Apparent sources of error, outside of variability in the kernels, are: Changes in the crushed kernels may not be the same as in the intact kernels, and there may be a passage of material other than water into or out of the kernel.

SUMMARY

Changes in the grain of corn silage may be determined under normal conditions of storage by using the kernel as a unit and applying statistical methods. Data from one silo show a loss of 5.08 per cent in the dry matter of the grain. The corresponding loss in crib storage was 2.25 per cent.

REVIEW OF FOREIGN DAIRY LITERATURE ¹

GORINI, COSTANTINO, Bacteriological Laboratory, Royal Superior School of Agriculture, Milan, Italy. Physiological Mutations of Bacteria by Means of Individual Divergencies.

The question of mutations is of the greatest importance to the geneticist. It is known that DeVrie's theory is in antithesis to the Darwinian doctrine insofar as it contends that the origin of species does not take place gradually, by adaptation and selection, but abruptly as a result of internal causes. Thus, to the Darwinian concept of continuous variability is to be replaced the concept of discontinuous variation. As a matter of fact there are arguments in favor of both these theories, and it is reasonable to assume that they may coexist.

Controversies have centered upon the definition of the term *Mutation*. The essential characters given by DeVries for this phenomenon are: sudden and spontaneous appearance, lack of finality and the fact that mutations are hereditarily fixed. Each of these characters has been the object of extensive discussion so that now amplifications and limitations have been applied in various cases. Leaving aside the botanical and zoological phases of the subject and viewing the matter solely from the bacteriological standpoint, it should be stated that we are here concerned with delicate and long series of observations which may well lead to errors and erroneous interpretations.

Neisser (1) and Massini (2) were the first authors to mention mutation among bacteria and they were soon followed by a host of imitators; it should nevertheless be mentioned that not all the workers have given sufficient proof of the purity of the cultures they were working with and of the fact, that the variations they observed were not due to external factors, ceasing with the return of these outside conditions.

In fact it is logical to overlook the spontaneity in appearance of the variation under consideration, and to consider as mutations also modifications of experimental origin provided their permanence can be proved after the removal of the causal experimental conditions. One eloquent example in this connection is furnished by the work of Schierbeck (3) who, working with lactic bacteria observed phenomena of attenuation which were constant even in continued transfers from

¹ This article was translated and presented for publication by Augusto Bonazzi, Wooster, Ohio.

milk to milk; so already in 1900, i.e., when mutations were not even spoken of as yet this author assumed he was working with "*lasting varieties*." Nevertheless when these cultures were transferred into special milk, very old and repeatedly sterilized, Schierbeck found them to regain their original zymogenic powers. He thereby concluded that ordinary milk had an inhibiting effect upon the lactic ferment and that the absence of this inhibiting factor in the special milk allowed these organisms to regain their original vigor. This is a typical example of such pseudomutations as very easily mislead the investigator who does not take into careful consideration such essential precautions as are dictated by the knowledge of the extreme sensitivity of bacteria towards even the slightest variations in experimental conditions.

Principal requisites of these investigations, says DeVries, are accuracy and perseverance.

In order to avoid the nearly inevitable errors in technique which easily creep in during these investigations, familiarity with the bacterial species under examination is necessary; a familiarity which can only be acquired after long periods of cultivation. I have transferred, as an example, my lacto-proteolytic organisms from time to time, and at the variable intervals indicated in my previous communications (4), for the last fifteen to twenty years.

It is by this means that I have been able to detect in every detail even slight differences in behavior towards temperature, aerobiosis, quality of nutritive media (especially milk), differences which lead at irregular intervals to irregular oscillations of the complex acido-coagulating and peptonizing activity. This irregularity I have for the past always considered of a transitory nature, and it is only recently that the repeated appearance of certain phenomena (5) which I had up to the present considered due to contamination or to inevitable accidents in the *modus operandi*, led me to ascertain true mutations characterized by the classical sudden appearance, spontaneity and independence from external factors and by their hereditary characters. This last condition is not to be taken in the absolute meaning of "*perennial constancy*;" it is sufficient that the new character be transmissible for a few generations. I have in fact observed also cases of reversibility, true *retromutations*, sudden and transmissible which DeVries himself partially admits.

I have been able to determine a new fact: that we are not dealing here with a transformation involving the whole of a bacterial culture, since not all the parallel transfers from the same mother culture yield the

modification but only some among them. This I have found to be the case although I took the precaution that the mother culture originate from a single cell obtained by the method of Burri.

In an attempt to find an explanation for the above mentioned phenomena I found it necessary to admit the possibility of an individual divergence in the cells of the same species of lacto-proteolytic ferments. An individuality which allowed the classification of the cells into three main types with regard to their saccharolytic and proteolytic properties; one type with these two properties in equilibrium, one other prevalently saccharolytic, the third preeminently proteolytic. According to this interpretation, mutations lose the appearance of abnormality and fall within the realm of normal phenomena subordinated only to the laws of chance (even though rare) whereby the material in the transferred inoculum is made up exclusively or almost exclusively of cells of the same type and falling only within one of the above groups. So that among the many transfers from the same mother culture there may exist side by side with the *mutant* cultures of the original type. In its turn again, the mutant may again at a given moment, and due to the play of chance, give rise on further transfer to a culture typical of the original variety (*retromutation*). Due to chance, is here again emphasized, under conditions of eugenesis and independently of disgenetic conditions as has been claimed by some authors (6). Therefore the possibility, should be admitted among bacteria of a cellular individuality similar to that which has been demonstrated for yeasts isolated by the single cell method, according to the method of Hansen.

Such considerations led me to take up again the study of several points of dairy bacteriology; to begin with my own personal researches.

In my first contribution upon the subject of the mammary flora (7) I listed five types of cocci which already at that time I would not attempt to consider as separate species but only as physiological types, since I found them to be distinctly linked by even ever-so-distant similarities.

In my other works upon the subject, I did not emphasize these distinctions but limited myself to the qualification of the mass of mammary cocci as lacto-proteolytic, since I was then convinced of the difficulties entailing a sharp differentiation of these organisms into peptonizing and non-peptonizing cocci. In fact I had been using as a criterion of the diversity of these five types their behavior in gelatine and milk, since I had at first noticed a certain similarity between the peptonizing powers on gelatine and that same power on casein. Nevertheless already at that time my attention had been drawn to the appearance

here and there of such types as I designated in my notes of the time, "*mixed types*" and which were to be classified in different groups according to whether they were considered with regard to their behavior in milk or gelatine.

Further studies (8) led me to call attention to the existence of cocci incapable of dissolving gelatine, but capable instead of dissolving casein, and still later I noticed the contrary phenomenon of gelatine-liquefying cocci which when placed in milk would coagulate but not peptonize the casein over long series of transfers. But now, benefiting by my later experience on the phenomena of mutation, I am in a position to state that if many subcultures are prepared from the same mother culture, liquefying or non-liquefying, discordant results may be obtained, i.e., daughter cultures which in some cases have simply coagulating and in other cases both coagulating and peptonizing properties.

A second group of organisms to which this line of reasoning, I thought, would be worth extending is that of the *Streptococcus lacticus*, an organism which according to my researches (9), now reconfirmed (10), is capable of peptonizing casein only at low temperatures of incubation. Also in this case I found that a single colony can yield transfers which when placed in milk are capable of different proteolytic powers, in some cases failing entirely in this property for some unknown reason and independently of the quality of the milk as well as of the experimental procedures. This together with other reasons pointed to in earlier publications, may explain why various authors failed to ascertain peptonizing properties in *Streptococcus lacticus*.

Observations of a similar nature I made on a third group of lacto-proteolytic bacteria of milk, i.e., *Lactobacillus sporificans*. This group is typified by the *B. acidificians-presamigenes-casei* which I described in 1904 (11) in cheese and which I considered as a first example of a spore-forming lactic ferment. Others were added to the list, some isolated from silage (12) and others from fermented milk products (13). A new type has*recently been isolated and described in butter by Sandelin (14).

In view of the above considerations it is now more appropriate to classify these three groups into variations closely associated with individual divergencies among cells of the same species rather than peptonizing or non-peptonizing species or races. These variations may be transitory, due to the *modus operandi* or other circumstances favoring either saccharolysis or proteolysis, and again there may be permanent and transmissible acquiring thereby the characters of mutations due to

the element of chance in the make up of the transfer inoculum as has been stated above.

How many of the properties of the lactic ferment are subject to these mutations due to simple individual divergencies rather than to adaptations or forced modifications? The Gram-staining properties may be mentioned in this connection properties which have been by some investigators assumed useful in the differentiation of true from pseudo-lactic ferments. Here again we may mention the classical example of *B. coli* so closely allied to the lactic bacteria, its fermentative affinities and its innumerable physiological races isolated, which have also been made to include the one of *B. coli* mutabile. Are these true races or cellular individualities within the same race?

These investigations lead me to caution against the eventual appearance of mutations in the selected cultures such as are used in the dairy. To avoid such mutations it is advisable to add to the scrupulous identity of developmental conditions, the caution of using abundant inoculi taken from the whole depth of the mother culture, since it should be considered possible that the various cellular types occupy different layers, or zones, in the growth.

Nevertheless my long experience has shown that these precautions are not in themselves sufficient, since, in spite of their constant adoption in my transfers, modifications have crept into my cultures for butter and for cheese manufacture. Although up to the present time I had leaned towards the assumption of weakenings or degenerations under unnatural conditions, I am now more inclined to consider these modifications as consequences of cellular individuality since I was able to show that even in these cases normal cultures may be obtained if the precaution be taken of making several parallel transfers.

Naturally, although mutations are not caused by environmental conditions, they are apt to be more sharply differentiated from the original type under unfavorable conditions. It is therefore to be recommended in making "*selected cultures*" to use media that will further a rapid development and to make several parallel transfers.

These phenomena of cellular individuality may be used in an attempt to overcome the difficulties attending the classification of the lactic bacteria even though the physiological criteria be used in preference to the morphological according to my findings (4).

A clear example of this is to be found in the intricate systematization, recently proposed by Orla Jensen, and based on cultural-biochemical complexes (15). It must be admitted that the attempt is laudable but

also that the results obtained fall far short of expectation and this due to the numerous uncertainties encountered. Taking as an example the criteria based upon the presence or absence of acido-proteolytic properties we find Jensen to confirm my previous work when he finds his property present in several of his lactic ferments, but I will also state that, had he not submitted his milk to the drastic sterilization which he recommends in his Treatise (16) he would have found this property far more widespread conforming to my findings purporting to show (17) that excessive sterilization of milk renders this substance unfit for the demonstration of caseolytic properties of the lactic bacteria. Now, Jensen having observed the presence of this property in some forms and its absence in others, makes of it a criterion for the differentiation of species.

From the standpoint of the action of these organisms on the carbohydrates, Jensen distinguishes species fermenting some sugars from others which never do so, as well as those which may or may not attack them according to circumstances. In an attempt to explain these inconsistencies he recognizes in the species, tendencies to undergo weakenings or degenerations. It is only natural if we advance the following question: Why do not the other species show these tendencies? If we admit the influence of cellular individualities, we find a plausible explanation for these phenomena, not only, but actually avoid the danger of creating new species and a new terminology not based on sufficiently sound criteria.

SUMMARY

In previous contributions I have shown that the group of the acido-proteolytic lactic ferments presents a great variability and irregularity in the manifestation of their coagulating and peptonizing properties according to external conditions as well as the *modus operandi* in experimental procedure.

In a recent note I have shown that they are capable of showing phenomena of true mutation, spontaneous, sudden and transmissible: having, nevertheless, found these mutations localized in portions of a culture I have recognized in them normal variations in cellular individuality.

In the present contributions I attempt to show that these considerations throw a new light upon a number of questions interesting dairy bacteriology: questions relating to the qualitative and quantitative irregularities to be observed in the activities of these organisms, dis-

tinguishable from errors of technique and observation, responsible for the endless and ever insufficient number of species, subspecies, races and varieties, created by investigators, as well as for the process of hypothetical attenuation or degeneration which do not find adequate justification in disgenetic conditions. These individual divergencies are also responsible for the ever increasing tendency towards the acceptance of the terminology *group* and *type* in place of *species*, contrary to the dignity of the science of bacteriology.

On the principle of normal cellular individuality we are in position not only to dispense with the necessity of explaining mutations and the conditions leading thereto, but also to simplify the general classification of the lactic ferments, contrary to the modern tendency inclining towards excessive complication.

These researches also find practical application in the culture, and selection of cultures, of lactic bacteria in the laboratory and the industry. In fact although the so-called mutations are inherent to the cell itself, not dependent upon external conditions or the *modus operandi*, we can easily see how their appearance will be propitiated by the use of small inoculi and by such conditions of culture as tend towards the enhancement of these "*normal*" cell divergencies. Therefore we must recognize the advisability of using large inoculi of taking these inoculi from all portions of the mother culture, and of making several parallel transfers and of using appropriate media such as milk which, according to my teachings, must be absolutely fresh and not drastically sterilized (tyndalized rather than autoclaved).

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TENNESSEE UNIVERSITY

Prof. C. E. Wylie of the Dairy Department reports the following news: Mr. Thomas B. Harrison, a graduate of Purdue University, has accepted the position of instructor in dairying at the University of Tennessee. Mr. Harrison will hold the position formerly held by Mr. Myron A. Loomis, now manager of the Jersey Farm and Milk Company, Nashville, Tenn.

The Dairy Department of the University of Tennessee is conveniently located on the first floor of the new \$265,000 Agricultural Building. Recently \$11,000 worth of new equipment has been installed for the activities of the department. All equipment in the creamery is new and modern and is operated by individual electric motors for each piece of machinery. The equipment includes a 5-ton refrigerator outfit, two 200-gallon pasteurizing vats, one 600-pound churn, scales, sterilizers, bottles, cans, etc. The creamery is operated on the commercial basis, buying cream for the manufacture of creamery butter, and buying milk for pasteurization and bottling for city supply.

CALIFORNIA UNIVERSITY

Prof. C. L. Roadhouse reports that the new Dairy Industry Building at Davis is nearly completed and will be ready for use at the opening of the school year in August.

MASSACHUSETTS AGRICULTURAL COLLEGE

Prof. W. P. B. Lockwood resigned as head of the Dairy Department, effective April 1. He has accepted a position as managing director of the New England Dairy and Food Council, with headquarters at 51 Cornhill, Boston, Mass. Professor Lockwood will, however, continue to spend about one-third of his time with the college as extension professor in dairying. Prof. H. F. Judkins has been appointed acting head of the dairy department.

J. H. F.

THE HEAT COAGULATION OF MILK¹

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In the sterilizing process in the manufacture of evaporated milk, it is desirable to produce an incipient coagulation, in the nature of a tender jell or "liver" as it is called by the practical men. Such a product will have a heavy creamy body after the "liver" has been broken up by the shaking process. This is desirable because the product presents a richer appearance to the consumer, and on account of the greater viscosity, there is little danger of fat separation from such a product.

However, evaporated milk differs widely in the readiness with which it coagulates. At times the milk coagulates so readily that it would be unmarketable, on account of its curdy appearance, if it were heated long enough to preserve it. This constitutes the problem with which we are concerned in this paper.

There is probably no condensery that is entirely free from this trouble throughout the year. It is undoubtedly one of the biggest problems that confronts the industry.

SUMMARY OF PREVIOUS ARTICLE

In a previous article (1) this problem was investigated indirectly in a study of the factors that influence the heat coagulation of fresh milk at 136°C. The following conclusions were based on this study:

1. In fresh milk there is no relation between the titratable acidity and the heat coagulation.
2. The hydrogen ion concentration of fresh milk is not the determining factor in the heat coagulation.

¹ Published with the permission of the Director of the Wisconsin Agricultural Experiment Station.

3. The concentration of the milk influences the heat coagulation, but it accounts only partly for the wide differences in the heat coagulation of fresh milk samples.

4. The main factor in the heat coagulation of fresh milk is the salt composition of the milk, especially the calcium, magnesium, citrate and phosphate content. The effect of the calcium and magnesium salts is opposed to the effect of the citrates and phosphates and vice versa. An excess of either of these two classes of salts causes the milk to coagulate more readily; the proper balance produces the most stable condition.

Since the publication of the previous paper the above conclusion on the importance of the milk salts has been substantiated by a study of a large number of samples of fresh milk from a number of different cows. Whenever a sample was found that coagulated at 137 to 138°C.² the effect of additions of sodium citrate, di-potassium phosphate, calcium acetate, and sodium bicarbonate was observed. Sodium bicarbonate was included because it is so commonly used in evaporated milk.

In all cases it was possible to prevent the coagulation by a small addition of salts normally present in milk. Table 1 gives a few typical results selected from a large number.

Table 1 gives the coagulation of the milk samples under the addition of the solutions of salts in amounts ranging from 0.0 to 0.4 cc. per 25 cc. of milk. In all cases the dilution was equalized by the addition of the proper amount of distilled water.

It will be noted from a study of table 1 that in sample 1 the addition of sodium citrate, sodium phosphate, and sodium bicarbonate prevented the coagulation. The addition of a soluble calcium salt hastened it. In the other four samples the opposite is true; the calcium acetate prevented the coagulation and sodium citrate, sodium phosphate and sodium bicarbonate either did not improve it or actually hastened it.

The action of the sodium bicarbonate is similar to that of the citrates and phosphates; it differs from them in that larger addi-

² The xylol used in the xylol vapor bath in the previous paper boiled at 136°C. In this paper the xylol used boiled at 137 to 138°C. This difference is due to differences in purity.

TABLE 1
The influence of salts on the heat coagulation of fresh milk

SAMPLES	2% CO. OF MILK PAYS:		Coagulation at 137°C. in minutes							
	{ ec. of water..... ec. of solution..... }		0 0	0 4	0 3	0 2	0 1	0 2	0 1	0 0
			0 0	0 0	0 1	0 2	0 3	0 2	0 3	0 4
Coagulation at 137°C. in minutes										
1. Clothilde, 11/20/20	Sodium citrate m/4.....	2:45	3:15	3:30	20:00—	20:00—	20:00—	20:00—	20:00—	6:00
	Sodium phosphate m/4.....	2:45	3:15	20:00—	20:00—	20:00—	20:00—	20:00—	20:00—	20:00—
	Calcium acetate m/4.....	3:00	3:15	3:00	1:00	0:30	0:15	0:15	0:15	0:15
	Sodium bicarbonate m/2.....	3:00	3:15	4:00	20:00—	20:00—	20:00—	20:00—	20:00—	20:00—
2. Wingers, 11/23/20	Sodium citrate m/4.....	2:00	2:00	2:00	1:45	1:45	1:45	1:45	1:45	1:45
	Sodium phosphate m/4.....	2:00	2:00	2:00	1:55	1:45	1:45	1:45	1:45	1:45
	Calcium acetate m/4.....	2:00	2:00	2:30	3:00	20:00—	20:00—	20:00—	20:00—	20:00—
	Sodium bicarbonate m/2.....	2:00	2:00	2:00	2:00	2:00	2:00	2:00	2:00	2:00
3. Lilac, 12/3/20	Sodium citrate m/4.....	3:30	3:30	2:00	2:00	1:45	1:45	1:45	1:45	1:45
	Sodium phosphate m/4.....	3:30	3:30	2:00	2:00	2:00	2:00	2:00	2:00	2:00
	Calcium acetate m/4.....	3:30	4:00	4:00	20:00—	20:00—	20:00—	20:00—	20:00—	20:00—
	Sodium bicarbonate m/2.....	3:30	4:00	1:15	1:30	1:30	1:30	1:30	1:30	10:00
4. Lady, 12/2/20	Sodium citrate m/4.....	2:45	2:45	2:45	2:45	2:30	2:30	2:30	2:30	2:30
	Sodium phosphate m/4.....	2:45	2:45	2:45	2:30	2:30	2:30	2:30	2:30	2:30
	Calcium acetate m/4.....	3:00	3:00	4:00	20:00—	20:00—	20:00—	20:00—	20:00—	20:00—
	Sodium bicarbonate m/2.....	3:00	3:00	2:00	2:00	2:00	2:00	2:00	2:00	20:00—
5. Lily, 11/24/20	Sodium citrate m/4.....	3:15	3:15	2:45	2:15	2:15	2:15	2:15	2:15	2:00
	Sodium phosphate m/4.....	3:15	3:15	2:30	2:30	2:30	2:30	2:30	2:30	2:30
	Calcium acetate m/4.....	3:15	3:15	20:00—	20:00—	20:00—	20:00—	20:00—	20:00—	4:00
	Sodium bicarbonate m/2.....	3:15	3:15	2:15	2:30	2:30	2:30	2:30	2:30	20:00—

tions prevented the coagulation even where smaller additions hastened it. This indicates that the influence of the sodium bicarbonate is twofold—it has a balancing effect on the calcium, and it changes the reaction. On this basis the results obtained with samples 2, 3, 4 and 5 can be explained as follows:

These samples coagulated because of a low content of calcium. The sodium bicarbonate tends to increase this deficiency by counteracting the effect of the calcium present; as a result the coagulation is hastened. However, with the larger additions of the sodium bicarbonate the reaction of the milk is changed to such an extent that the casein is held in solution, the sodium probably replacing part of the calcium in the casein.

EVAPORATED MILK

The underlying idea in the work on the heat coagulation of fresh milk was that the factors that influence the coagulation of fresh milk would also be applicable to evaporated milk. The demonstration that the salts which are normally present in milk have a very decided effect on the stability of the casein in fresh milk is quite conclusive. This logically leads to the conclusion that they must have a similar effect on the evaporated milk, for we are here dealing with an entirely similar system, modified only to the extent to which it has been concentrated and the heat treatment and other manipulations it has undergone. The experiments carried on with evaporated milk and the observations made under commercial conditions verify this conclusion.

On account of the small amounts of the normal milk salts that affect the heat coagulation, their effect on evaporated milk was studied by adding them to the milk, using small amounts well within the limits of normal variations in milk. A number of such experiments were made on evaporated milk produced under commercial conditions. In most cases the salt solutions were added to the milk in the can, the can sealed, shaken and immediately sterilized. In several cases their effect was studied when added before the milk was concentrated. The results are given in tables 2, 3, 4 and 5.

TABLE 2

Evaporated milk from condensery A, December 29, 1920

16 OUNCES OF EVAPORATED MILK PLUS:	cc. of water.....	0 0	12 0	9.0	6.0	3.0	0.0
	cc. of solution.....	0.0	0.0	3.0	6.0	9.0	12.0
Calcium acetate M/4.....	3*	3	3+	4	5	6	
Sodium phosphate M/4.....	3	3	2	2-	1	1	
Sodium bicarbonate M/2.....	3	3	3-	2+	2	1	

* Indicates the relative firmness of the coagulum.

TABLE 3

Evaporated milk from condensery B, December 29, 1920

8 OUNCES OF EVAPORATED MILK PLUS:	cc. of water.....	0.0	4.0	2.0	0.0
	cc. of solution	0.0	0.0	0.2	0.4
Sodium citrate M/4.....	1*	1	1	2	
Calcium acetate M/4.....	1	1	4	5	
Sodium phosphate M/4.....	1	1	2	3	
Sodium bicarbonate M/2.....	1	1	3	4	

* The figures indicate the relative firmness of the coagulum.

TABLE 4

Evaporated milk from condensery C, March 5, 1921

8 OUNCES OF EVAPORATED MILK PLUS:	cc. of water.....	0 0	3 0	2.5	2.0	1.5	1.0	0 5	0.0
	cc. of solution..	0 0	0.0	0 5	1.0	1 5	2.0	2.5	3 0
Sodium phosphate M/4....	2S*	1S	0+S	0+R	0+R	0+R	0+R	0+R	0+R
Sodium citrate M/4.....	2S	1S	1S	0+S		0+S	0S	0S	
Calcium acetate M/4.....	2S	1S	2S	3S	3+S	4R	4+R	5R	
Sodium bicarbonate M/2...	2S	1S	0S	0+S	0+S	1+S	1+S	2S	

* The numerals indicate the relative firmness of the coagulum. S indicates that the body of the milk was smooth on shaking. R indicates that the body of the milk was rough on shaking.

TABLE 5

Evaporated milk from condensery D, March 13, 1921

16 OUNCES OF EVAPORATED MILK PLUS	cc. of water.....	0.0	6.0	4.5	3.0	1.5	0.0
	cc. of solution...	0.0	0.0	1.5	3.0	4.5	6.0
Sodium citrate M/4.....	2S*	2-S	1+S	1S	1-S	1-S	
Potassium phosphate M/4.....	2S	2-S	2-R	1+R	1R+	1-R++	
Calcium acetate M/4.....	2S	2-S	3R	4R	5R+	6 (whey)	
Sodium bicarbonate M/2.....	2S	2-S	1+R	1+S	1-S	1S	

* Same as in table 4.

The results in tables 2, 3, 4 and 5 show that the addition of sodium citrate, sodium phosphate, calcium acetate and sodium bicarbonate has the same effect on the evaporated milk as on the fresh milk and casein solutions. The observations in table 2 were made in an attempt to increase the process at a condensery that had considerable trouble with the coagulation. The results showed that both sodium phosphate and bicarbonate improved the milk. Sodium bicarbonate added to the milk greatly increases the caramelization and impairs the flavor, as a result its further use was not advisable. Sodium citrate had not been tried in this case but its effect always is similar to that of the phosphate, and since we had only sodium citrate on hand in sufficient amounts, its use was recommended in this case. Two ounces of sodium citrate were added per 1000 pounds of raw milk, and this increased the sterilizing process four minutes at 240°F. This addition which amounted to an increase of 0.0067 per cent in the citric acid content of the milk changed the sterilizing process from an unsafe to a satisfactory process.

The results in tables 3, 4 and 5 were gathered merely as a demonstration; they had no immediate practical application since the evaporated milk in all three cases was entirely satisfactory without any addition. The fact that sodium bicarbonate, which is so commonly used, can have an injurious effect on the process is shown in tables 3 and 4.

The effect of calcium chloride on the coagulation of the evaporated milk was accidentally demonstrated at condensery D when a small amount of calcium chloride brine leaked into the milk at the aerater. This small amount of calcium chloride, not enough so that the analysis of the milk showed an abnormal content of calcium, caused the milk to coagulate so severely even at the minimum sterilizing process that it would not flow out of the open can when it was inverted. By adding the proper amount of sodium citrate, within the limits of normal variation of this constituent in milk, and heating up the milk by drawing it through the vacuum pan, the product finally processed at 244°F. for sixteen minutes. It was entirely normal in appearance, taste and analyses. This was a striking illustration on a large

scale (50,000 pounds of milk) that the salts have a very marked effect on the coagulation of the evaporated milk.

The beneficial effect of sodium citrate and di-sodium phosphate has been demonstrated in a number of other plants, according to reports from condenseries that had trouble with low sterilizing processes, and where the use of these salts had been recommended. One company operating several condenseries writes,

We have been trying out the di-sodium phosphate with marked success. In some instances it has enabled us to extend our (sterilizing) process four or five minutes and it has consistently given us two or three minutes of extra time. . . . We have saved ourselves a great deal of trouble by using this method.

Thus it is demonstrated that the salt balance is of importance in the manufacture of evaporated milk. The process of adjusting the salt balance in order to avoid low sterilizing processes is already an accepted commercial practice in a number of plants as a result of this study.

The conclusion that the milk salts have a decided effect on the heat coagulation of evaporated milk is contrary to the conclusion of Rogers, Deysher and Evans (2). In their study of the factors involved in the coagulation of evaporated milk they concluded that there is no definite relation between the "acid-base ratio" and the coagulation and that the composition of the milk salts is "only a minor factor in determining the coagulating temperature of evaporated milk." As their criterion in arriving at this conclusion they applied the analytical method used by us in our previous paper. The milk samples were analyzed for calcium, magnesium, citric acid and phosphoric acid, and the excess of calcium and magnesium in gram equivalents over the citric acid and phosphoric acid was calculated. Using this method as a criterion the samples having the largest excess of calcium and magnesium should coagulate most readily. Rogers, Deysher and Evans failed to find this relation and based their conclusion entirely on this fact.

Their conclusion undoubtedly is erroneous because of the inadequacy of the analytical method which they used as their criterion. In our previous paper this method was applied to

fresh milk not as the sole test but simply as an interesting analytical verification of a fact which had already been amply demonstrated synthetically. We found only a general agreement between the excess of calcium and magnesium and the coagulability of fresh milk. There are several considerations which can readily account for any irregularities:

1. The analyses deal with the total content of the salts while only that portion of the salt remaining in solution as the temperature is raised affects the coagulation.

2. The amounts of the salts necessary to affect the coagulation are very small, within or approaching the limits of experimental error of the analytical methods.

3. The calculations are based only on approximate results as to the equivalency of the salts.

4. Other factors such as concentration and reaction are not taken into consideration.

These considerations which cause only a general instead of a detailed agreement between the calculated excess of calcium and magnesium and the coagulation of fresh milk, become of even greater moment in attempting to apply this calculation to evaporated milk. The extent to which the salts have been changed to an insoluble form is greater in evaporated milk, and the reaction of evaporated milk is more acid so that the equivalency of the salts may be different. As a result it is not surprising to find that this method used as the sole criterion leads to erroneous conclusions.

THE ACTION OF THE MILK SALTS IN THE COAGULATION

It has been suggested that the addition of the salts changes the reaction of the milk and that the action of the salts in preventing the coagulation is to be attributed to this rather than to a direct influence of the salts on the stability of the casein.

Zoller (3) in working with solutions of milk salts found that the solutions were more acid in reaction after heating and that the change in reaction was proportional to the amount of calcium precipitated. He found further that the amount of calcium

precipitated and the change in acidity was greatly reduced by doubling the citrate content of the salt solutions. The conclusion from this work appears to be that the beneficial effect of sodium citrate in preventing the heat coagulation of milk is to be attributed to its indirect effect on the reaction. That this explanation is inadequate is demonstrated by the following experiments.

The change in reaction caused by the addition of a salt to the milk may be considered in three different ways:

1. The salt added to the milk at room temperature may change the reaction due to the alkalinity or acidity of the salt or to its buffer effect.

2. If the milk is heated after the salt has been added there may be a change in reaction attributable to the salt, due to interaction of the milk salts and precipitation of calcium phosphate.

3. The reaction may be considered in a dynamic sense. It is a well-known fact that the dissociation constant of water varies very decidedly with the temperature. In a similar manner the dissociation constants of the various milk salts vary with the temperature. The final reaction that results in the milk at any given temperature is determined by the equilibrium which obtains between all the constituents, each with its characteristic dissociation constant. As a result there is the possibility that two samples of milk of the same reaction at room temperature, but of a different composition, may have reactions differing from each other at higher temperatures. In a complex substance such as milk it is impossible to calculate what this change in reaction with a given change in temperature will be. However, it is probable that there may be an appreciable difference in the reaction of two milk samples having the same reaction at room temperature.

That the beneficial effect of the addition of a salt to the milk is not to be attributed to a change in reaction in the first and second sense as outlined above is demonstrated by the following experiments (tables 6 and 7).

A sample of heat coagulable milk from an individual cow of the University herd was selected. The reaction of this milk was

studied both before and after the addition of calcium acetate in sufficient amounts to prevent the coagulation. The results are given in table 6.

The coagulation of both samples in table 6 was prevented by the addition of calcium acetate. The results show that there was little or no change in the reaction accompanying the addition of the salt to the milk at room temperature, so that the beneficial effect cannot be attributed to a change in reaction in this sense. After the samples had been heated at 100°C. for twenty minutes the samples that had the calcium acetate added were slightly more acid than those that were heated similarly without the calcium acetate present. However, the difference was so slight

TABLE 6

The change in reaction accompanying the addition of calcium acetate to the milk

	625 CC. OF MILK PLUS:			
	Sample 1, February 15, 1921		Sample 2, March 1, 1921	
	cc. of distilled water	cc. of calcium acetate	cc. of distilled water	cc. of calcium acetate
Heat coagulation at 137°C.....	2:30	20:00—	2:30	20:00—
Reaction at room temperature, pH.....	6.59	6.59	6.60	6.58
Reaction at room temperature after heating at 100°C. for twenty minutes, pH..	6.53	6.48	6.51	6.48
Change in reaction due to heating pH..	0.06	0.11	0.09	0.10

that very little importance can be attached to it. As will be shown further in table 7 the beneficial effect of the calcium acetate cannot be attributed to this slight increase in acidity.

Sample 2 was one that was actually benefited by an increase in acidity but the increase necessary was much greater than that obtained in the case of adding calcium acetate. This is shown by the following experiment: The sample of milk was divided into two portions, one was kept at a temperature near the freezing point, and the other portion was placed in the incubator and the acidity allowed to develop to 0.65 per cent. A series of samples of varying acidity were prepared by mixing the two portions in varying proportions. The coagulation of the samples was studied

and the reaction of two samples determined. The results are given in table 7.

The results given in table 7 show that in this sample an increase of acidity prevented the coagulation, but the reaction at which this prevented the coagulation was much more acid than the reaction obtained under the optimum addition of calcium acetate as shown in table 6. This indicates that the calcium acetate (see table 6) had an effect as such and not by virtue of changing the reaction.

TABLE 7
Heat coagulation prevented by the increase in acidity

TUBE NUMBER	9 CC. OF MILK* (ACIDITY 0.15 PER CENT) PLUS SOUR MILK (ACIDITY 0.65 PER CENT)	ACIDITY OF MIXTURE LACTIC ACID	COAGULATION AT 137°C.	pH
	<i>cc. of sour milk</i>	<i>per cent</i>		
1	0.0	0.150	2:30	6.60
2	0.1	0.155	5:00	
3	0.2	0.160	6:00	
4	0.3	0.166	20:00—	
5	0.4	0.171	20:00—	6.23
6	0.5	0.176	20:00—	
7	0.6	0.182	20:00—	
8	0.7	0.187	0:45	
9	0.8	0.193	0:30	
10	0.9	0.198	0:15	

* This is the same sample of milk as sample 2 in table 6.

The question of whether or not the action of the salts in preventing the heat coagulation is to be explained by their influence on the reaction in the dynamic sense cannot be answered directly, for it is impossible to measure the reaction at the high temperature at which the heat coagulation takes place. However, the question can be answered indirectly by demonstrating that the milk salts have a very decided effect on the stability of the casein under somewhat different conditions.

It has been shown by Van Slyke and Hart (4) and by Van Slyke and Bosworth (5) that calcium chloride has a marked effect on the solubility of casein in lime water. Their work showed

that in neutralizing a lime water solution of casein with hydrochloric acid the neutralization could be carried much farther without the precipitation of the casein if the calcium chloride formed was removed by dialysis. The calcium chloride has the effect of precipitating the casein.

If the citrates and phosphates counteract the effect of the calcium in the heat coagulation, then they should show a similar action in preventing the precipitation of the casein at room temperature. As a preliminary experiment to demonstrate this, 4 grams of purified casein were dissolved in 100 cc. of 0.9737 $N/20$ calcium hydroxide solution. To portions of this solution hydrochloric acid, sodium phosphate, and sodium citrate were added at room temperature. The results are given in table 8.

From a study of table 8 it is seen that the citrates and phosphates counteract the effect of the soluble calcium salts on the precipitation of the casein. If the soluble calcium salts are properly balanced with the citrates or phosphates then the casein is held in solution even in an acid reaction (trials 6, 9, 12, 13 and 14). In the absence of citrates and phosphates the casein is precipitated even in alkaline solutions (trials 3 and 4). In trial 3 the addition of calcium acetate to the solution of casein in limewater causes the partial precipitation of the casein. In trial 4 the small amount of calcium chloride formed early in the neutralization with hydrochloric acid causes the partial precipitation of the casein.

If now we neutralize the excess of lime water of the casein solution by means of an $N/10$ hydrochloric acid solution that contains dissolved in it enough citrate to make it also a $M/20$ sodium citrate solution, we then shall always have a molecule of sodium citrate present for every molecule of calcium chloride that is formed in the neutralization.

To demonstrate that the sodium citrate does not keep the casein in solution by affecting the reaction, a solution of casein in limewater was neutralized by means of the hydrochloric acid-sodium citrate solution with vigorous stirring until the casein just started to precipitate out. The reaction of the solution was found to be pH 4.81. This demonstrates conclusively that the

TABLE 8
The effect of milk salts on the precipitation of casein from solution

TRIAL NUMBER	10 CC. OF CASEIN SOLUTION PLUS:	RESULT	REACTION TO LITMUS
1	1 cc. sodium phosphate $m/4$	Solution remains clear	Alkaline
2	1 cc. sodium citrate $m/4$	Solution remains clear	Alkaline
3	1 cc. calcium acetate $m/4$	Cloudy—casein precipitates	Alkaline
4	2 cc. HCl $N/10$	Cloudy—casein precipitates	Alkaline
5	No. 4 plus 1 cc. Na Citrate $m/4$	Solution clears up	Alkaline
6	No. 5 plus 2 cc. HCl $N/10$	Solution remains clear	Acid
7	1 cc. Na citrate $m/4$, 1 cc. Ca acetate $m/4$ plus 2 cc. HCl $N/10$	Solution very milky	Alkaline
8	No. 7 plus 1 cc. Na Citrate $m/4$	Solution clears up	Alkaline
9	No. 8 plus 2 cc. HCl $N/10$	Solution remains clear	Acid
10	No. 3 plus 1 cc. Na citrate $m/4$	Casein redissolves	Alkaline
11	No. 4 plus 1 cc. Na_2HPO_4 $m/4$	Casein redissolves	Alkaline
12	1 cc. Na_2HPO_4 $m/4$ and 4 cc. HCl $N/10$	Solution remains clear	Acid
13	No. 12 plus 1 cc. Ca acetate $m/4$	Casein precipitates	Acid
14	No. 13 plus 1 cc. Na_2HPO_4 $m/4$	Solution clears up	Acid

sodium citrate has an effect as such and does not keep the casein in solution by its effect on the reaction. The reaction was actually changed to pH 4.81, very close to the isoelectric point for casein before any precipitation took place.

The above experiments with casein solutions dealt entirely with their stability at room temperature. To demonstrate the effect of the sodium citrate on the stability of the casein in the heat coagulation, two series of casein lime water solutions were prepared. One series was neutralized to various stages with $N/10$ hydrochloric acid, the other series with one $N/10$ hydrochloric acid— $M/20$ sodium citrate solution. The dilution in all the samples in the two series was equalized by the addition of distilled water. The samples were subjected to the heat test, and from

TABLE 9

The effect of sodium citrate on the heat coagulation of a casein-lime water solution

25 cc. OF CASEIN SOLUTION PLUS:			HEAT COAGULA- TION	REACTION pH
Acid solution		cc. of water		
Kind	Amount			
	cc.			
N/10 HCl.....	1.0	7.0	0:10*	7.64
N/10 HCl, M/20 sodium citrate.....	8.0	0.0	20:00—	5.48

* Instantly.

each series the proper sample selected for the determination of the hydrogen-ion concentration. The results are given in table 9.

The casein solution in table 9 did not have the sodium citrate present coagulated instantly although its reaction was pH 7.64, while the solution that had the sodium citrate present did not coagulate at 137°C . in twenty minutes at a reaction of pH 5.48. With such a wide difference in the reaction and the behavior of the samples toward heat, we must attribute a direct effect to the sodium citrate that accomplished this great difference.

The effect of the normal milk salts on the coagulation is further emphasized by a study of the effect of sodium citrate and calcium chloride on the coagulation of the casein dissolved in sodium hydroxide (table 10), the effect of sodium phosphate and calcium

TABLE 10
The effect of sodium citrate and calcium chloride on the coagulation of casein solutions

cc. $\frac{M}{4}$ sodium citrate	5 cc. OF CASEIN SOLUTION PLUS:										
	$\frac{M}{4}$ calcium chloride										
	0.0	0.1	0.2	0.3	0.4	0.5	0.6	0.7	0.8	0.9	1.0
0.0	—	x	1:00	0:40 xx	0:30 xx	0:20 xx	xxx	xxx	xxx	xxx	xxx
0.1	—	—	x	1:00	0:50 xx	0:30 xx	0:20 xx	xxx	xxx	xxx	xxx
0.2	—	—	—	x	0:30	0:30 xx	0:20 xx	xxx	xxx	xxx	xxx
0.3	—	—	—	—	x	0:40	0:30 xx	0:25 xx	xxx	xxx	xxx
0.4	—	—	—	—	—	5:00	0:25	0:20 xx	xxx	xxx	xxx
0.5	—	—	—	—	—	3:30	3:00	0:25	0:15 xx	xxx	xxx
0.6	—	—	—	—	—	3:30	3:00	1:00	0:25	0:15 xx	xxx
0.7	—	—	—	—	—	3:30	1:30	1:20	0:50	0:30	0:15 xx
0.8	—	—	—	—	—	3:30	1:30	1:20	1:00	0:50	0:40
0.9	—	—	—	—	—	4:00	1:30	1:20	1:00	0:50	0:40
1.0	—	—	—	—	—	20:00	2:00	1:00	0:50	0:40	0:30

* The casein solution consists of 40 grams of purified casein dissolved in 1 liter of $\frac{N}{10}$ NaOH, and then neutralized with 700 cc. of 1.042 $\frac{N}{10}$ NCl, to a reaction of pH 6.60. (The dilution was equalized in all cases with water.)

--, Clear after heating at 137°C. for twenty minutes.

x, Cloudy after heating at 137°C. for twenty minutes.

xx, Cloudy at room temperature before heating.

xxx, Coagulated at room temperature before heating.

chloride on the coagulation of the sodium hydroxide-casein solution (table 11), and the effect of sodium citrate, sodium phosphate and calcium acetate on the coagulation of the casein-lime-water solution (table 12). The results obtained with these solutions where the conditions are less complex than in the milk, are entirely in accord with those obtained with the milk itself.

TABLE 11

The effect of sodium phosphate and calcium chloride on the coagulation of casein solution

5 cc. of casein solution* plus:											
CC. M/4 POTAS- SIUM PHOS- PHATE	CC. M/4 CALCIUM CHLORIDE										
	0 0	0.1	0.2	0.3	0.4	0.5	0.6	0.7	0.8	0.9	1.0
0.0	--	--	xx	xxx	xxx	xxx	xxx	xxx	xxx	xxx	xxx
0.1	--	--	--	xxx	xxx	xxx	xxx	xxx	xxx	xxx	xxx
0.2	--	--	--	--	xxx	xxx	xxx	xxx	xxx	xxx	xxx
0.3	--	--	--	--	0:10	xxx	xxx	xxx	xxx	xxx	xxx
0.4	--	--	--	--	--	0:10	xxx	xxx	xxx	xxx	xxx
0.5	--	--	--	--	--	--	xxx	xxx	xxx	xxx	xxx
0.6	--	--	--	--	--	--	xxx	xxx	xxx	xxx	xxx
0.7	--	--	--	--	--	--	xxx	xxx	xxx	xxx	xxx
0.8	--	--	--	--	--	--	0:10	xxx	xxx	xxx	xxx
0.9	--	--	--	--	--	--	--	0:10	xxx	xxx	xxx
1.0	--	--	--	--	--	--	--	--	0:10	xxx	xxx

* The casein solution consists of 40 grams of purified casein dissolved in 1 liter of N/10 NaOH, and then neutralized with 700 cc. of 1.042 N/10 NCl, to a reaction of pH 6.60. (The dilution in each case is equalized by the addition of water.)

xxx, See table 10.

xx, See table 10.

--, See table 10.

If in tables 10 and 11 we to draw a diagonal from the upper left to the lower right hand corner and divide the table each into two fields, we find that in the upper where the calcium chloride is in excess the coagulation takes place most readily, and in the lower field where the sodium citrate (table 10) and the Phosphate (table 11) are in excess the casein solutions do not coagulate at all. The diagonal should be the boundary between these two extremes. With the larger additions of sodium citrate and cal-

cium chloride in table 10 the experimental results deviate from this expected result. The explanation probably is that the higher concentration itself has an effect on the coagulation. In

TABLE 12

The effect of sodium citrate, potassium phosphate, and calcium acetate on the coagulation of casein-lime-water solution

	20 CC. OF CASEIN SOLUTION* PLUS:			HEAT COAGULATION AT 137°C.
	cc. of 5 per cent Na citrate	cc. of 5 per cent Ca acetate	cc. of water	
Sodium citrate calcium ace- tate balance	0.00	0.00	0.00	20:00—
	0.75	0.00	0.50	20:00—†
	0.75	0.10	0.40	20:00—†
	0.75	0.20	0.30	20:00—
	0.75	0.30	0.20	20:00—
	0.75	0.40	0.10	2:00
	0.75	0.50	0.00	0.15
	cc. of 5 per cent K ₂ HPO ₄	Ca acetate cc. 5 per cent	cc. of water	HEAT COAGULATION AT 137°C.
Di-potassium phosphate calcium acetate balance	0.00	0.00	0.00	20:00—
	1.25	0.00	3.75	18:00‡
	1.25	0.10	3.65	19:00‡
	1.25	0.20	3.55	20:00‡
	1.25	0.30	3.45	20:00‡
	1.25	0.40	3.35	20:00‡
	1.25	0.50	3.25	20:00‡
	1.25	0.60	3.15	0.30
	1.25	0.70	3.05	0.15
	1.25	0.80	2.95	0.15

* The casein solution consisted of 20 grams of purified casein dissolved in 500 cc. of 0.98738 N/20 lime water, neutralized to a reaction of pH 6.68 with 80 cc. of N/10-HCl-M/20 Na citrate and diluted to 800 cc.

† Granular precipitate.

‡ These samples did not show a pronounced precipitate until they were cooled; the precipitate seemed to form immediately then. Similar results were obtained when this experiment was repeated.

table 11 the results are quite sharp, either a sample did not coagulate at all or else it coagulated at room temperature. In tables 10 and 11 where we are dealing with casein-sodium hydroxide solutions, the excess of citrate or phosphate did not cause a

coagulation as they do in milk, but in table 12 where we are dealing with casein-lime water solutions the citrate and phosphate in excess caused a coagulation similar to that caused in milk by their excess.

Although these salts have a very decided effect on the coagulation of the evaporated milk when they are added to the milk after it has been concentrated, the result obtained in that manner are probably not fully indicative of what their effect would have been if they had been normally present in the milk or added before it was concentrated. A more thorough distribution is insured and the interaction of the salts is favored if they are present during the entire process of manufacture. It is common knowledge in the industry that forewarming to a high temperature or holding the milk longer at the high temperature has a beneficial effect in preventing the coagulation. This is undoubtedly due to the interaction of the salts, especially the precipitation of calcium, which is favored by this treatment. If the addition of a salt is to have its fullest effect it must be added to the milk before it is concentrated.

The results obtained under commercial conditions at condensery C will illustrate this, although the observations could not be made under as carefully controlled conditions as would be desirable. For a period of over a month starting early in June, the milk at this condensery coagulated very readily. Careful grading of the milk and rejecting the poor milk, did not remedy the trouble. Sodium bicarbonate only improved the milk slightly. The trials with the additions of sodium citrate and the sodium phosphate to the evaporated milk immediately before it was sterilized showed little improvement. However, the milk already had 3 ounces of sodium bicarbonate added per 1000 pounds of raw milk. The sterilizing process was twelve minutes at 240°F. In order to give the addition of sodium phosphate a fair trial, the use of the soda was discontinued and instead 1 ounce of sodium phosphate added per 1000 pounds of raw milk. This increased the sterilizing process from twelve minutes as it had been the previous day to sixteen minutes at 240°F. There had been no change in the weather conditions and the process had been around

twelve minutes at 240°F. consistently for a number of days, so that it is quite safe to assume that the improvement was due to the difference of adding 1 ounce of sodium phosphate instead of 3 ounces of soda. There is the possibility that the mere elimination of the soda may have been beneficial, but repeated tests had shown that more soda could have been added without injurious effect. While several factors might have contributed to this change the indication is that the sodium phosphate when added to the milk at the forwarmers showed an appreciable improvement, where it had shown little improvement when added to the milk after it had been concentrated.

SUMMARY AND CONCLUSIONS

1. The main factor in the heat coagulation of fresh milk is the salt composition of the milk.
2. The salt composition of the milk is an important factor in the heat coagulation of evaporated milk.
3. In a number of cases the troublesome coagulation of evaporated milk has been remedied on a commercial scale by the addition of the proper amount of sodium citrate or di-sodium phosphate.
4. The milk salts affect the coagulation of milk, both fresh and evaporated, directly. Their effect is not indirect through a change in reaction.

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THE SEASONAL VARIATIONS OF THE PER CENT OF FAT IN COW'S MILK

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The relation between the season of the year and the percentage of fat in milk was shown by Eckles (1) from a study of 240 lactation periods of cows in the Missouri and Iowa Experiment Station herds. He found that regardless of when the lactation began, the per cent of fat when plotted followed a general curve for the year, being lowest during June and July and gradually rising to the highest point in December and January and then again declining until mid-summer. The curves were found to be modified by the evident tendency for the percentage of fat to rise materially during the last two or three months of the milking period. White (2) found that the same relation exists between the solids-not-fat. From a study of the records of the Pacific Creamery and of cows in thirty herds in cow testing associations in the Salt River Valley, Arizona, Clothier (3) concluded that the most probable cause of the difference in fat content of milk between the winter and summer months was the difference in the nature of the feed.

The object of this paper is to present additional data showing the seasonal variations of the per cent of fat in cow's milk.

The data presented in the tables and charts were derived from a study of 3763 Guernsey, 299 Jersey and 95 Holstein-Friesian records.

The Guernsey records were secured from the Advanced Register of Guernsey Cattle. The animals used were representative with respect to their ability as producers except as the minimum requirement of the Advanced Register excludes low producers. The animals were scattered over the United States to some extent but a majority of the records were made in the east and

central western states. These records, therefore, were made under varying conditions from the standpoint of climate, feed, and management.

The Jersey records consist of Register of Merit tests of Jersey cattle made in Missouri during the past few years. Animals of varying producing ability (except those unable to meet the minimum requirements of the Register of Merit) are represented. In this case climatic conditions were similar, being restricted to one state.

The Holstein-Friesian records were secured from the Advanced Register and private records of the Holstein-Friesian herd owned by the University of Missouri. Animals of widely varying productive ability are represented since all cows completing yearly records were included. All cows were under similar herd conditions.

It should be noted that, although varying from year to year, these data include separate records of three breeds made under varying climatic conditions. The Guernsey data is of a national character; the Jersey data is confined to a single state (Missouri); while the Holstein-Friesian data is confined to a single herd.

The records of each breed were divided into groups according to the month during which the lactation began, i.e., all cows calving in January were placed together, those in February together, etc. Records made during parts of two lactation periods were eliminated. The average per cent of fat for each month was obtained by totalling the percentages of fat and dividing this total by the number of cows in the group. This method was used rather than the so-called "true average" per cent obtained by dividing the total of the fat by the total of the milk due to the fact that the first method is used by the various breed associations in obtaining the average per cent of fat when several separate tests are made on a cow during a single month. To be consistent the same method was used in our work. A comparison was made of the two methods, however, and quite similar results were obtained.

By grouping the records according to the month during which the lactation began the influence of the stage of lactation as a

factor in the variation of the per cent of fat in the milk can be shown in conjunction with seasonal variations. In a previous paper (4) we presented values derived from the same data showing the influence of the advance of lactation on the per cent of fat. Theoretically, if there were no other factors affecting the per cent of fat in cow's milk during a lactation period, the values obtained for the average per cent of fat by each group beginning the lactation period during the various months would follow the same general curve as the values derived without consideration of the month when the lactation began. A significant difference was found when these two values were plotted together.

The data for the Guernsey breed is presented in table 1 and chart 1. The continuous lines in the charts are the plotted values showing the influence of the stage of lactation on the percentage of fat (4) while the columns are the plotted values obtained for the cows grouped according to the month during which the lactation began showing the percentage of fat throughout the lactation period.

In the groups starting their lactation period during January, February, March and April, it will be seen that the percentages of fat during the summer months are considerably below the average figures for the advance of lactation. However, in September there is a rapid increase in the per cent of fat approaching very closely the average figures. During the rest of the lactation the percentages of fat exceed the average figures.

The extremely high percentages of fat during the winter months is again shown very strikingly in the groups whose lactation periods began during May, June, July and August. Starting in September and October the percentages of fat rise above the average until the following spring (April or May) when tests again tend to fall off in spite of the well known tendency for the percentage of fat to increase rapidly during the last stages of the lactation period.

In the groups beginning their lactation periods during October, November and December the seasonal variations in the per cent of fat is clearly shown. In these groups the decrease in the percentages of fat due to the season of the year occur during the

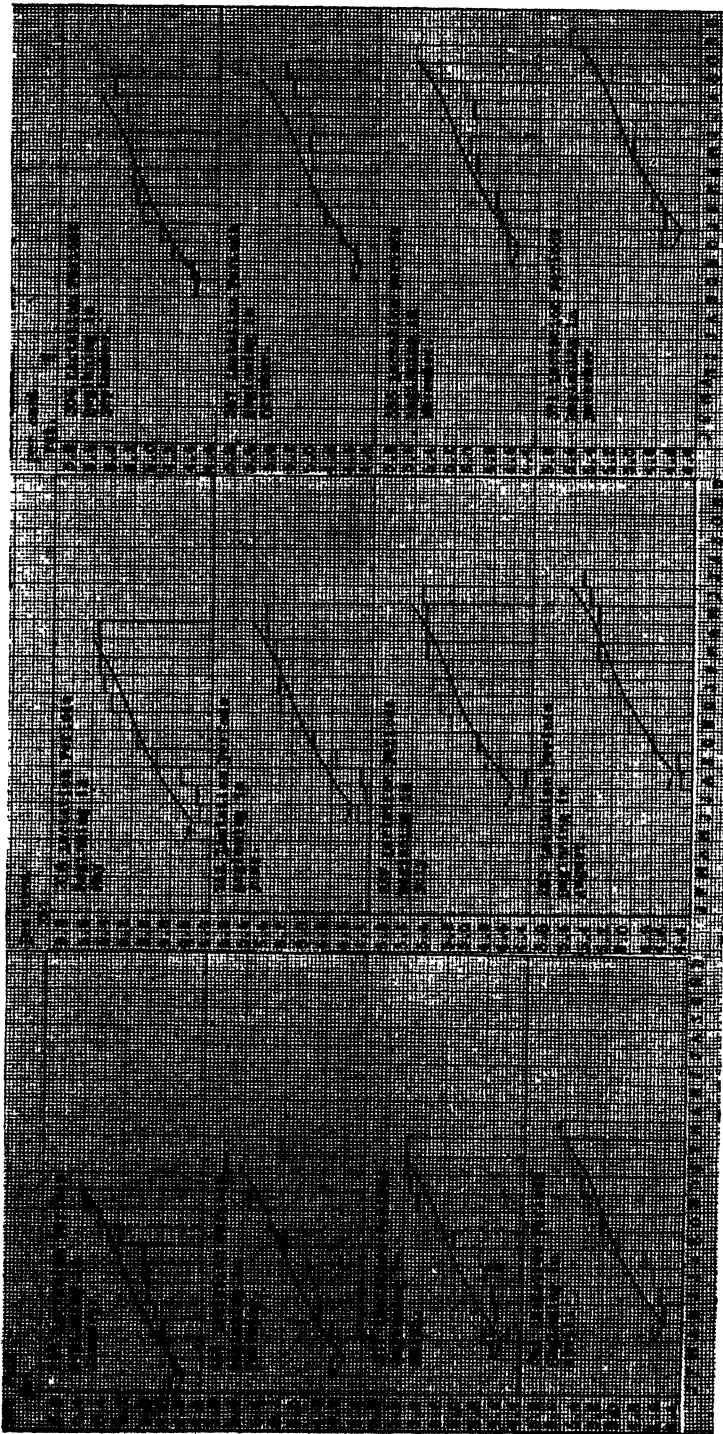


CHART I. INFLUENCE OF THE SEASON OF THE YEAR ON THE PER CENT OF FAT IN COW'S MILK. GUERSEY

Continuous lines show influence of the stage of lactation on the percentage of fat.

Columns show seasonal variations in percentage of fat for cows grouped according to the month in which the lactation began.

latter part of the lactation period and show conclusively that the season of the year exerts a greater influence upon the percentage of fat than does the advance of lactation.

TABLE 1
Influence of the season of the year on the per cent of fat in cow's milk
Guernseys

	NUMBER OF COWS CALVING DURING											
	January 297	February 267	March 377	April 361	May 318	June 242	July 237	August 225	September 306	October 367	November 395	December 371
January.....	4.70											
February.....	4.64	4.70										
March.....	4.72	4.60	4.67									
April.....	4.79	4.70	4.55	4.65								
May.....	4.85	4.81	4.64	4.57	4.60							
June.....	4.92	4.82	4.72	4.61	4.55	4.54						
July..	4.92	4.88	4.76	4.68	4.57	4.45	4.48					
August.....	4.98	4.97	4.83	4.77	4.65	4.51	4.43	4.49				
September ..	5.23	5.18	5.10	5.04	4.95	4.76	4.73	4.53	4.62			
October.....	5.45	5.37	5.28	5.22	5.16	4.90	4.95	4.79	4.65	4.66		
November ..	5.58	5.56	5.45	5.37	5.34	5.20	5.20	5.03	4.83	4.64	4.68	
December.....	5.72	5.73	5.54	5.43	5.41	5.32	5.32	5.14	4.99	4.79	4.70	4.78
January.....		5.78	5.58	5.52	5.48	5.34	5.40	5.23	5.14	4.93	4.78	4.74
February..		5.68	5.55	5.54	5.32	5.39	5.39	5.30	5.21	5.04	4.81	4.84
March....			5.64	5.55	5.38	5.44	5.33	5.22	5.08	5.00	4.94	
April				5.58	5.44	5.48	5.31	5.20	5.07	5.01	5.00	
May.....					5.49	5.46	5.32	5.23	5.08	5.07	5.05	
June						5.51	5.42	5.23	5.07	5.02	5.03	
July.....							5.49	5.31	5.12	5.06	4.92	
August							*	5.45	5.24	5.15	5.09	
September..									5.36	5.40	5.39	
October.....										5.64	5.58	
November.....											5.71	

The height of the column above or below the continuous line is an index of the extent of the seasonal variation of the per cent of fat. For example with the group of cows starting their lactation period in March the per cent of fat in July is 0.21 lower than the average figure. The opposite effect is best shown with the group of cows starting their lactation period in July in which

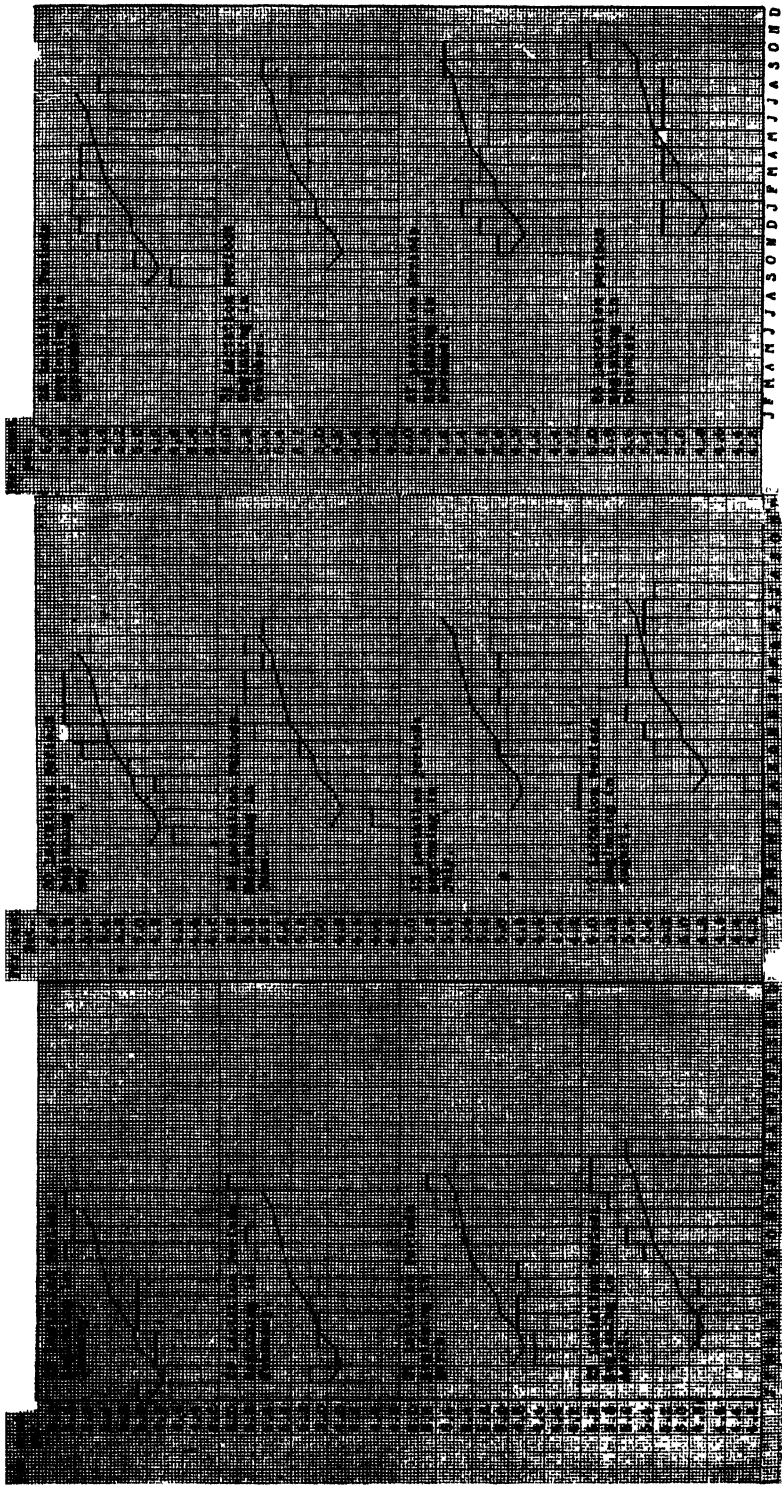


CHART II. INFLUENCE OF THE SEASON OF THE YEAR ON THE PER CENT OF FAT IN COW'S MILK. JERSEY

Continuous lines show influence of the stage of lactation on the percentage of fat.

Columns show seasonal variations in percentage of fat for cows grouped according to the month in which the lactation began.

case the per cent of fat in January is 0.34 higher than the average figures. In general it may be said that no matter when the lactation period begins, there is a tendency for the per cent of fat to rise above the average during the winter months especially

TABLE 2

*Influence of the season of the year on the per cent of fat in cow's milk
Jerseys*

	NUMBER OF COWS CALVING DURING											
	January 28	February 25	March 37	April 29	May 30	June 20	July 13	August 7	September 24	October 31	November 27	December 28
January.. .. .	5.11											
February.....	4.80	5.02										
March.....	4.99	4.99	4.72									
April.....	4.99	5.15	4.71	4.83								
May.....	5.06	5.19	4.90	4.93	4.73							
June.....	5.12	5.27	4.99	5.07	4.93	4.50						
July.....	5.11	5.15	4.82	4.93	4.87	4.60	4.46					
August.....	5.03	5.20	4.96	5.14	4.97	4.80	4.46	4.43				
September.....	5.90	5.80	5.21	5.59	5.27	4.60	4.85	5.00	4.75			
October.....	5.68	5.96	5.57	5.72	5.77	5.35	5.15	5.43	5.17	5.00		
November.....	5.78	5.87	5.62	5.86	5.80	5.80	5.08	5.57	5.50	5.00	5.11	
December.....	5.88	6.07	5.82	5.97	5.93	6.00	5.08	5.71	5.75	5.26	5.30	5.32
January.....		6.10	5.90	6.03	5.93	5.90	5.15	6.00	5.83	5.32	5.48	5.29
February.....			5.61	6.10	5.90	5.90	5.08	5.71	5.88	5.39	5.41	5.21
March.....				5.79	5.80	5.70	5.15	5.71	5.71	5.19	5.37	5.39
April.....					5.60	5.90	5.23	5.71	5.61	5.22	5.37	5.36
May.....						5.70	5.23	5.57	5.42	5.22	5.19	5.43
June.....							5.31	5.57	5.46	5.22	5.22	5.36
July.....								5.43	5.42	5.03	5.19	5.29
August.....									5.50	5.39	5.15	5.32
September.....										5.77	5.70	6.04
October.....											5.67	6.11
November.....												6.00

December, January, and February and then to gradually decline during the spring and summer reaching the lowest point during June, July and August. These seasonal variations have been shown by Ragsdale and Brody (5) to be due to environmental temperature.

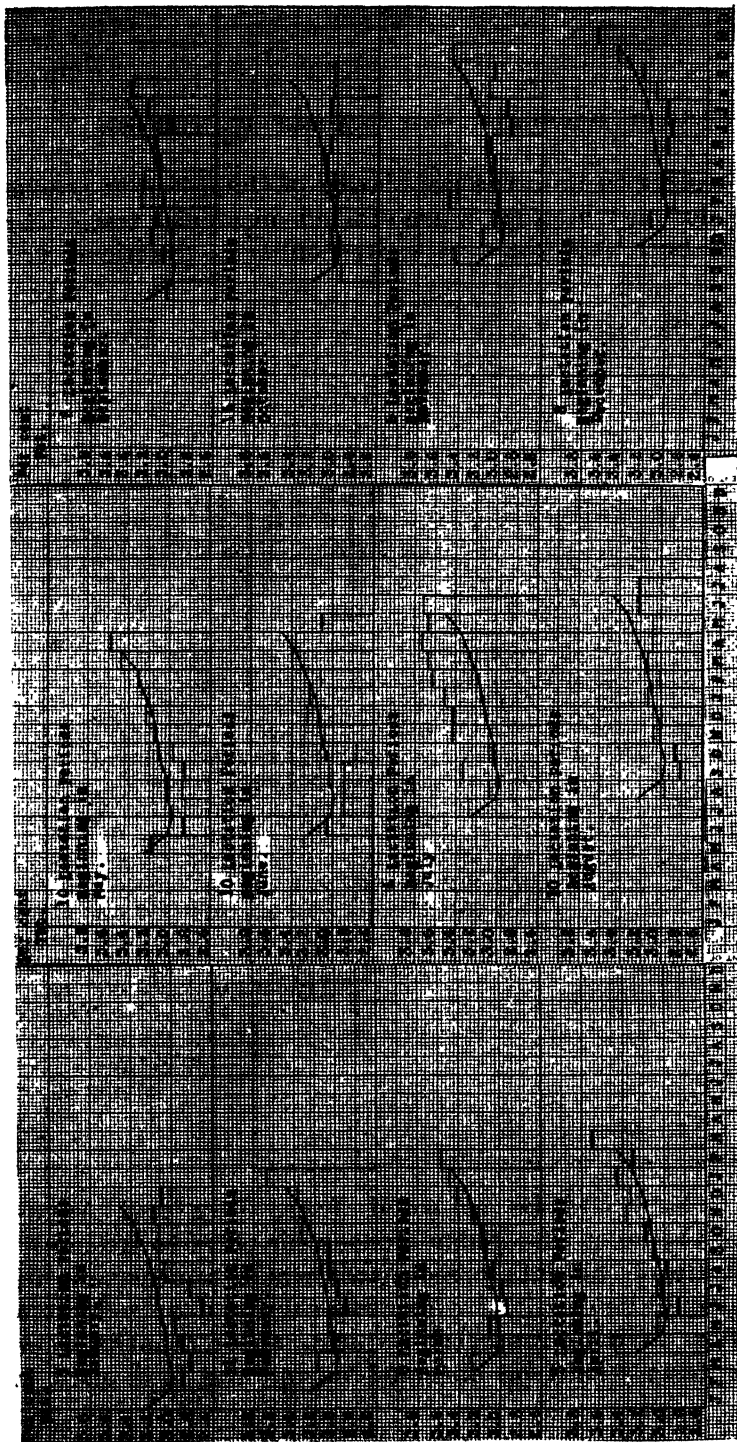


CHART III. INFLUENCE OF THE SEASON OF THE YEAR ON THE PER CENT OF FAT IN COW'S MILK. HOLSTEIN

Continuous lines show influence of the stage of lactation on the percentage of fat.

Columns show seasonal variations in percentage of fat for cows grouped according to the month in which the lactation began.

It is interesting to note the popularity of the different seasons of the year during which cows are started on official test. It is evident that both fall and spring freshening is practiced very

TABLE 3
Influence of the season of the year on the per cent of fat in cow's milk
Holsteins

	NUMBER OF COWS CALVING DURING											
	January 9	February 11	March 8	April 3	May 10	June 10	July 6	August 10	September 8	October 6	November 6	December 8
January.....	3.20											
February.....	2.87	3.53										
March.....	2.93	3.19	3.29									
April.....	2.89	3.03	2.98	3.48								
May.....	2.97	3.09	3.09	3.08	3.19							
June.....	2.71	2.90	2.93	2.86	2.87	3.05						
July.....	2.82	2.99	2.98	3.08	2.85	2.88	3.29					
August.....	3.07	3.05	3.07	2.99	2.87	2.88	3.08	2.98				
September.....	3.12	3.05	3.00	3.14	2.86	2.88	3.34	2.81	3.08			
October.....	3.12	3.34	3.18	3.20	2.97	2.76	3.08	2.87	3.01	2.95		
November.....	3.18	3.33	3.26	3.38	3.16	3.07	3.42	3.08	3.10	3.01	3.44	
December.....	3.11	3.38	3.41	3.35	3.23	3.25	3.41	3.00	3.22	3.06	3.18	3.41
January.....		3.67	3.49	3.44	3.20	3.23	3.50	3.14	3.17	3.03	3.07	2.16
February.....			3.58	3.27	3.31	3.12	3.62	3.01	3.32	3.00	3.55	2.94
March.....				3.71	3.47	3.40	3.66	3.15	3.25	3.05	3.04	3.03
April.....					3.59	3.37	3.73	3.07	3.14	3.11	3.05	3.01
May.....						3.11	3.66	3.07	3.19	3.06	3.10	2.99
June.....							3.71	3.23	3.03	2.96	2.89	2.88
July.....								3.22	3.25	3.06	2.91	2.98
August.....									3.42	3.03	3.11	3.10
September.....										3.04	3.09	3.21
October.....											3.48	3.48
November.....												3.67

extensively. As would be expected June, July and August appear to be the most unpopular months for cows to freshen.

From a study of the data presented it would appear that the best results would be obtained with cows calving during September or October. With cows calving during these months somewhat higher tests would be obtained during the first eight or

nine months of the lactation period when the cows are producing a maximum amount of milk.

The data for the Jersey breed is presented in table 2 and chart 2. Due to the fact that fewer records were available, as compared to the Guernsey breed, the plotted curves are not as smooth and some irregularities appear. Still the trend is the same. The groups of cows whose lactations begin in January, February and March show the decline in the percentage of fat during the summer months as compared to the average percentages of fat also the increase in the percentage of fat beginning in September and continuing during the winter months is apparent.

The decline in the percentage of fat during the following summer months is clearly shown with those groups whose lactation periods begin in September, October and November in spite of the advance of lactation.

The data for the Holstein-Friesian breed is presented in table 3 and chart 3. Since in most of the groups the number of cows was limited the seasonal variations of the percentages of fat is not as clearly shown. In the larger groups, however, the same trend is apparent as in the other breeds.

SUMMARY

Data was presented showing seasonal variations of the percentage of fat in cow's milk derived from a study of 3763 Guernsey, 299 Jersey, and 95 Holstein-Friesian yearly records.

The percentage of fat in milk when plotted follows a general curve, being lowest during the summer months, then gradually rising reaching a peak during the winter months and then again declining during the spring and summer.

When the different seasons of the year are accompanied by varying temperatures, such as ordinarily prevail throughout the greater portion of the United States, the influence upon the per cent of fat in cow's milk is greater than that of the advance of lactation.

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A METHOD FOR THE QUANTITATIVE DETERMINATION OF GELATIN IN ICE CREAMS¹

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Since the present tentative method² of the Association of Official Agricultural Chemists for the detection of gelatin in milk states that a precipitate will be obtained in the presence of any considerable amount of gelatin, while smaller amounts will be indicated by a cloudiness, the implication is evident that an indication of the amount of gelatin present can be gained by observing the appearance of the precipitate.

To find out to what extent the method could be made quantitative, the turbidity produced by the addition of picric acid as directed was compared with that formed when a standard gelatin solution was treated in exactly the same way. To get a turbidity that could be well observed in the nephelometer, it was found necessary that the filtrate, after the removal of the proteins with acid mercuric nitrate, should contain from 0.025 to 0.005 per cent of gelatin. The filtrate prepared according to the tentative method was diluted until an equal volume of saturated picric acid solution gave a turbidity approximating that produced by treating similarly a 0.01 per cent solution of gelatin which had been previously tested and found to be of the highest quality. This standard gelatin solution should contain acid mercuric nitrate in as nearly as possible the same amount as the unknown solution. The results shown in table 1 were obtained by this procedure on 11 samples of ice cream mix prepared in the laboratory and containing known amounts of gelatin.

The filtrates obtained after the removal of the milk proteins with acid mercuric nitrate were practically clear when the

¹ Published by permission of the Department of Agriculture.

² Official and Tentative Methods of Analysis, page 229, Method No. 19.

samples contained small amounts of gelatin and were decidedly cloudy when 1 per cent or more of gelatin was present. The percentages of gelatin in the experimental ice cream (table 1) were low when 0.40 to 0.50 per cent of gelatin was present in the original sample, but were high when the samples originally contained larger amounts. A good approximation of the amount present, however, can be made by this procedure which has the advantage of being short.

TABLE 1
Estimation of gelatin in experimental ice cream by use of nephelometer

SAMPLE NUMBER	COMPOSITION OF SAMPLE			GELATIN FOUND
	Fat	Sugar	Gelatin	
	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>	<i>per cent</i>
1	17.0	11	0.40	0.30
2	14.0	11	0.40	0.30
3	9.0	11	0.40	0.37
4	14.0	15	0.47	0.31
5	8.5	14	0.50	0.36
6	9.5		0.50	0.35
7	14.0	15	0.75	0.85
8	13.0	15	0.99	1.33
9	14.0	10	1.01	1.27
10	12.0	15	1.27	1.63
11	13.0	10	1.31	1.61

A more accurate determination of gelatin in ice cream can be made by precipitating the casein with acetic acid, throwing out the gelatin with alcohol, and dissolving the gelatin in hot water. The gelatin recovered in this way is practically free from milk protein and gives a clear solution that can be read in the polariscope for rotation and mutarotation as determined by C. R. Smith.^{3,4} The detailed procedure is as follows:

PROCEDURE A

Weigh 450 grams of ice cream into a 500-cc. flask, warm to 35°, and dilute to the mark. Transfer to a larger flask and add

³ Jour. Amer. Chem. Soc., xli (1919), 135.

⁴ Jour. Ind. Eng. Chem., xii (1920), 878.

10 per cent acetic acid until the casein is coagulated, observing the volume of acetic acid added. Calculate the volume of the fat and casein (volume of fat = weight of fat in sample \times 1.075; volume of casein = weight of casein in sample \times 0.73, and determine the volume of serum by subtracting this from 500 plus the number of cc. of acetic acid added. If the curd does not settle readily, shake with a small amount of carbon tetrachloride, centrifuge, and decant the supernatant serum. Measure 100 cc. of the serum into a 400-cc. lipped beaker and while still warm add slowly, with stirring, 200 cc. of 95 per cent alcohol. Cool in ice water and let stand until the precipitate settles, leaving the supernatant liquid clear. At this point the precipitate may be allowed to settle over night in an ice box at as near zero Centigrade as possible. Filter on a large hardened paper in a Buchner funnel and wash with cold alcohol, two volumes of 95 per cent alcohol and one of water. Drain the precipitate, remove to a small beaker, and rub up with 50 cc. of water. Allow to soak at room temperature until the gelatin swells, then heat to about 90°C. in a hot water bath, filter, and wash with hot water. Make up to 100 cc. at 35° and polarize. Cool a portion of the solution rapidly to 15°, pour into a cold polariscope tube, let stand at 15° for twenty-four hours, and polarize again. The reading at 15°, divided by the reading at 35°, will give an indication of the jelly strength of the gelatin.⁴ Calculate the percentage of gelatin in the sample from the reading at 35° from the following formula:

$$\text{Percentage of gelatin} = \frac{R \times 0.346 \times V \times 100}{W \times 2 \times \text{specific rotation}}$$

When W = weight of sample; V = volume of acetic acid serum; and R = Polariscopes reading in Ventzke degrees in 2 decimeter tube.

The specific rotation of high-grade gelatin is given by Smith⁵ as 141, when calculated to a moisture- and ash-free basis. Determine nitrogen in an aliquot of the gelatin solution and calculate the percentage of gelatin in the ice cream as follows:

$$\text{Percentage of gelatin} = \frac{N \times V \times 5.55 \times 100}{a \times W}$$

When W = weight of sample; V = volume of acetic acid serum; a = volume of aliquot of gelatin solution; and N = weight of nitrogen in aliquot of gelatin solution.

PROCEDURE B

When it is inconvenient to dilute the sample of ice cream to a definite volume and make a correction for the fat and casein, the following alternative procedure is proposed: Determine the reducing sugars in the ice cream according to the A. O. A. C. method for lactose in sweetened condensed milk.⁵ Weigh 450 grams of the ice cream in a beaker, warm to 35°C., and add 10 per cent acetic acid until the casein is precipitated. Mix thoroughly and filter or separate the serum by centrifuging. Neutralize a 5-cc. aliquot of the serum, dilute to 50 cc., determine the reducing sugars, and calculate the total volume of the serum by dividing the weight of reducing sugars in the 450-gram sample by the weight of reducing sugars in 1 cc. of the serum. To 100 cc. of the serum add 200 cc. of alcohol and continue as outlined under procedure A.

To show that comparable results for the volume of serum were obtainable by both procedures three samples of ice cream were treated according to procedure A. Reducing sugars were determined on the ice cream and on the serum prepared under procedure A, and the volume of serum was calculated by subtracting from the total volume the volume of the fat and casein and also by dividing the weight of reducing sugars in the entire sample by the reducing sugars in 1 cc. of the serum. These results are given in table 2. The difference found in each case was 2 cc., 1.8 cc., and 1.6 cc., which is sufficiently close for practical purposes of this work.

In order to show that gelatin could be quantitatively recovered by the procedure outlined, 21 samples of ice cream mix were prepared in the laboratory and analyzed. Varying percentages of fat, milk solids not fat, sugar, and gelatin were added as shown in

⁵ Official and Tentative Methods of Analysis of Association of Official Agricultural Chemists, p. 231, method no. 36.

table 4. The polariscopic observations on the samples of gelatin used in these experiments are recorded in table 3. These findings

TABLE 2
Difference between methods of calculating volume of serum

	1	2	3
Composition of ice cream mix { Fat, per cent.	40.3	14.1	13.1
{ Milk solids not fat, per cent.		6.0	6.8
{ Sugar, per cent.	14.0	14.0	14.0
{ Gelatin, per cent.	1.00	1.00	1.00
Lactose in sample, grams.	2.17	3.81	3.53
Lactose in 1 cc. of serum, grams.	0.0362	0.0446	0.0409
Calculated volume of solution (procedure B)	59.9	85.4	86.3
Volume of solution by correcting for volume precipitate (procedure A)	57.9	83.6	84.7
Difference.	2.0	1.8	1.6

TABLE 3
Data on samples of gelatin added to ice cream

SAMPLE NUMBER	NITROGEN	GELATIN	VENTEKE READING		READING AT 15° READING AT 35°	SPECIFIC ROTATION AT 35°
			35°	15°		
	<i>per cent</i>	<i>grams per 100 cc.</i>				
A	15.7	0.50	3.6	7.2	2.00	124.6
		0.75	5.4	11.4	2.11	124.5
		0.75	5.5	11.2	2.04	126.9
		1.00	7.1	14.54	2.05	122.8
B	14.9	0.20	1.4	2.5	1.79	121.1
		0.40	2.9	5.5	1.90	125.4
		0.50	3.52	6.8	1.93	121.8
		0.60	4.08	8.3	2.03	117.6
		0.75	5.15	10.3	2.00	118.8
		0.75	5.07	10.36	2.04	116.9
		1.00	6.80	13.75	2.02	117.6
C	15.7	0.75	5.3	8.1	1.53	122.3
		1.00	6.95	11.23	1.62	120.2
D	15.7	0.50	3.45	5.10	1.48	119.4

indicate that samples A and B of gelatin are high grade in jelly strength, while sample C is poorer and sample D is the poorest,

TABLE 4
Analyses of experimental ice cream

SAMPLE NUM- BER	GELA- TIN NUM- BER	COMPOSITION OF ICE CREAM MIX				WEIGHT OF SAMP- LES grams	VOLUME OF ACETIC ACID SERUM	VENTKEE READING*			NITRO- GEN IN GELA- TIN REC OV- ERED	GELATIN FOUND BY									
		Fat	Milk solids not fat	Sugar	Gela- tin			35°	15°	Reading at 15° Reading at 35°		Ventake at 35°	Nitrogen determi- nation	Specific rotation = 122°							
															per cent	per cent	per cent	per cent	grams	per cent	per cent
1†	A	3.2	7.6	11.0	0.50	300	290.6	3.70	6.70	1.81	0.0772	0.50	0.48	0.51							
								3.73	6.85	1.84	0.0772	0.50	0.48	0.51							
						300	299.6	3.75	6.82	1.82	0.0716	0.52	0.46	0.53							
								3.50	6.50	1.86	0.0716	0.49	0.46	0.50							
2	A	3.1	8.1	11.0	0.50	270	291.2	3.35	7.05	2.14	0.0760	0.50	0.52	0.51							
								3.30			0.0800	0.49	0.54	0.50							
3	A	3.2	7.6	11.0	0.75	300	291.0	5.20	10.40	2.00	0.1195	0.70	0.74	0.72							
								4.10	7.90	1.93	0.0913	0.73	0.74	0.74							
							291.0	5.05	9.40	1.86	0.1150	0.70	0.73	0.70							
								5.15	9.40	1.82	0.1150	0.72	0.73	0.71							
4	A	3.2	7.6	11.0	0.75	300	300.0	5.30	11.00	2.08	0.1195	0.74	0.76	0.75							
								5.25	11.00	2.09	0.1180	0.73	0.75	0.74							
								4.95	10.00	2.02	0.1130	0.73	0.76	0.74							
5	A	8.3		14.25	0.75	450	477.4	5.05	10.30	2.04	0.1125	0.74	0.76	0.76							
								5.10	10.20	2.00	0.1135	0.74	0.76	0.76							
6	A	9.1	8.1	14.0	0.75	450	472.2	5.00	10.10	2.02	0.1165	0.73	0.78	0.74							

7†	A	9.0	7.5	14.0	0.75	270	282.8	5.20 5.30	10.50 10.80	2.02 2.04	0.1165 0.1195	0.76 0.77	0.78 0.80	0.77 0.79
8†	B	3.1	7.5	11.0	0.20	300	290.9	1.40 1.30	2.50 2.50	1.79 1.92	0.0295 0.0308	0.19 0.18	0.19 0.20	0.19 0.18
9†	B	3.1	7.3	11.0	0.40	300	291.2	2.85 2.75	5.60 5.30	1.97 1.93	0.0590 0.0590	0.38 0.37	0.38 0.38	0.39 0.38
10	B	3.3	6.9	11.0	0.50	500	500.0	5.60 5.60	11.80 11.90	2.11 2.13	0.1400 0.1400	0.40 0.40	0.47 0.47	0.40 0.40
11	B	3.5	7.5	11.0	0.60	300	288.6	4.40	8.80	2.00	0.0913	0.61	0.59	0.60
12	B	3.1	7.6	11.0	0.75	300	300.0	4.85 4.85	9.50 9.30	1.96 1.92	0.1080 0.72	0.72 0.72	0.72	0.69 0.69
13	B	14.6	11.90	11.25	0.75	450	440.7	5.20 5.20	10.50 10.30	2.02 1.98	0.1214 0.1228	0.74 0.74	0.80 0.81	0.72 0.72
14	B	12.2	9.40	11.0	1.00	100	94.5	2.80	5.50	1.96	0.0618	0.95	0.98	0.94
15	B	3.3	8.80	11.0	1.00	300	300.0	6.37 6.40	12.90 13.00	2.02 2.03	0.1460 0.1445	0.90 0.91	0.98 0.97	0.90 0.91

* All readings are negative, the sign being purposely omitted.

† The alcohol precipitate was dissolved in water and again precipitated before making up to volume.

‡ Contained approximately 0.25 per cent Indian gum.

TABLE 4—Continued

SAMPLE NUM- BER	GELA- TIN NUM- BER	COMPOSITION OF ICE CREAM MIX				WEIGHT OF SAL- TIN FLUO- RES	VOLUME OF ACETIC ACID SERUM	VENTZKE READING*				NITRO- GEN IN GELA- TIN RECov- ERED	GELATIN FOUND BY		
		Milk solids not fat		Sugar	Gela- tin			35°	15°	Reading at 15° Reading at 35°	Ventzke at 35°		Nitrogen determi- nation	Specific rotation = 122°	
		per cent	per cent												per cent
16	C	7.4	6.70	14.80	0.75	453	481.3	4.50	7.50	1.67	0.0985	0.68	0.67	0.68	
								4.60	7.60	1.65	0.1025	0.69	0.70	0.69	
								4.50	7.50	1.67	0.1055	0.68	0.72	0.68	
17	C	7.9	7.40	11.0	1.00	450	478.8	6.30	10.10	1.60	0.1390	0.96	0.94	0.95	
								6.25	10.60	1.70	0.1420	0.96	0.96	0.94	
18†	D	8.8	7.70	15.0	0.50	450	474.5	2.40	3.50	1.46	0.0575	0.36	0.39	0.36	
								2.40	3.60	1.50	0.0535	0.36	0.36	0.36	
								2.20	3.50	1.59	0.0550	0.33	0.37	0.33	
19	D	8.0	8.10	15.0	0.50	450	477.7	2.65	4.00	1.51	0.0660	0.40	0.45	0.40	
								2.55	3.95	1.55	0.0630	0.38	0.43	0.38	
								2.80	4.10	1.46	0.0660	0.42	0.45	0.42	
20	D	8.0	8.10	15.0	0.50	450	479.7	2.40	3.50	1.46	0.0534	0.37	0.36	0.36	
								2.60	3.95	1.52	0.0618	0.40	0.42	0.39	
21	D	3.0	8.10	15.0	0.50	270	301.5	2.50	3.80	1.52	0.0575	0.41	0.41	0.40	
								2.40	3.65	1.52	0.0575	0.39	0.41	0.38	

* All readings are negative, the sign being purposely omitted.

† The alcohol precipitate was dissolved in water and again precipitated before making up to volume.

as was in fact the case. In each of the experiments reported in table 4 the volume of solution was determined by making up to a definite volume and correcting for the volume of fat and casein. The results were calculated from the Ventzke reading at 35° , using as a factor the values for each gelatin given in table 3 and again taking the average specific rotation as 122° and from the percentage of nitrogen in each sample of gelatin. The specific rotation 122° is merely the average experimental findings on samples employed and is lower than the specific rotation 141° given by Smith⁶ because of the presence of moisture and ash. In other words, using the specific rotation 122° in the calculation in place of 141° gives results for gelatin on the air-dried sample. The greatest difference between the result obtained by using the observed rotation of each gelatin and the corresponding result using 122° as the average specific rotation was 0.03 per cent. In each case the percentage of gelatin calculated from the nitrogen determination was very close to that obtained from the specific rotation. Table 4 also shows the ratio of the Ventzke reading 15° to that at 35° on the gelatin recovered from the ice cream. Since these values agree with the results given in table 3, it is evident that by this method the percentage of gelatin in ice cream can be determined and its jelly strength also measured.

In all these experiments, with the exception of no. 7, gelatin was the only thickening agent used. In experiment 7 some gum appeared to be mechanically removed with the casein. The percentage of gelatin found was very slightly higher than the amount added, showing that the quantity of gum present in this experiment had little effect upon the estimation of gelatin. The effect of gum and other ice cream thickeners upon the determination of gelatin by this method remains for future investigation. Since some gums are laevo-rotatory and some dextro-rotatory and may be precipitated with alcohol, however, their absence⁷ should be assured before interpreting the Ventzke reading obtained by the foregoing procedure as being due entirely to gelatin. To show that the reducing sugars have been completely

⁶ Loc. cit.

⁷ U. S. Dept. Agr., Bu. Chem. Bull. 116, p. 24.

removed by washing the gelatin precipitate with the dilute alcohol, their absence can be proved by testing the final gelatin solution with Fehling's solution. The presence of gelatin should be qualitatively determined by the picric acid test before making a quantitative determination by the polariscopic method.

SUMMARY

By diluting the filtrate, after removing the casein and fat with acid mercuric nitrate, according to the A. O. A. C. method for the detection of gelatin in milk, and comparing the turbidity produced by picric acid with a standard, the amount of gelatin in ice cream can be approximated.

More accurate results are obtained by precipitating the casein and fat with acetic acid, separating the gelatin with alcohol, and redissolving it in hot water. In this way the percentage of gelatin can be calculated from the nitrogen content and from the specific rotation of gelatin at 35° . The mutarotation can also be measured to show the jelly strength of the gelatin added to the ice cream. Results are shown on 21 samples of experimental ice cream mix, containing from 0.2 to 1 per cent of gelatin, using gelatins of good, medium, and poor quality.

A METABOLISM CRATE FOR CALVES AND OTHER SMALL RUMINANTS

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The metabolism crate described here is for use with calves and other small ruminants. It is felt that it will be satisfactory with sheep and goats as well as with calves as the only alterations necessary will be the grating on which the animal stands. It is also of low construction cost and easily portable. It is convenient to use in digestion and balance trials where the gaseous metabolism has not to be taken into consideration.

It consists of lower and upper parts as shown in figure 1 and there are two wheels on each side so that the upper half may be run off or on the lower section. The lower section has corner posts 4 inches square and these can be mounted on small wheels to render the moving of the crate easy.

This lower section is braced as shown in figure 1 and there is a board along each side so that it is easy to examine the animals. The collecting funnel, for the urine, of galvanized iron or zinc is attached to the lower part and the details are shown in figure 2. Running around the top of the lower section there is a 5-inch plank 1 inch thick and the funnel is so constructed that it fits into this and then runs back 4 inches on the top of the board. On top of this is a 5-inch plank, 1 inch thick which fits with the outer edge of the 6-inch plank. This 5-inch board is covered with galvanized iron on top and on the inner edge and is soldered around at this lower edge. On top of this are two runners 1 inch square to act as guards for the wheels attached to the upper section. One is set square with the outer edge of the platform and the other about $1\frac{1}{2}$ inches further in to allow room for the wheels which should be about $1\frac{1}{4}$ inches wide. The wheels are 3 inches in diameter and set in the lower part of the upper section and this gives plenty of room for them to work.

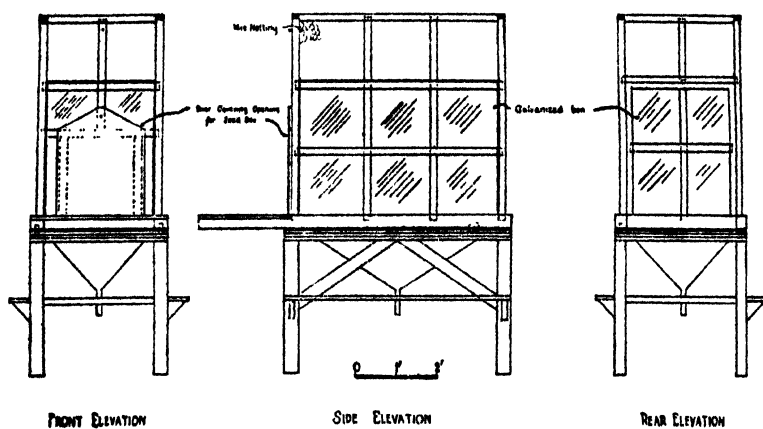


FIGURE 1

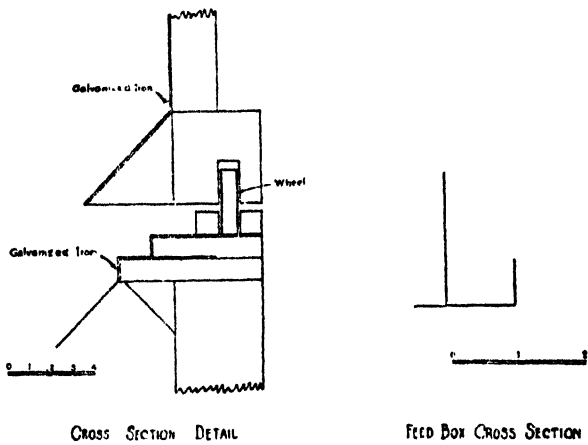


FIGURE 2

The base of the upper section, figure 2, consists of material 4 inches square and nailed to that on the inside is a quarter-sawed section of the same material. In front the basal material from the sides is carried from 2 to 3 feet forward of the crate proper, the top inch of this is taken out and platform of 1 inch material made on which to place the feed box and other material. The uprights, crossbars and top of the upper section are made of material 1 inch square as shown in figure 1. In the rear is inserted a door which reaches up to the level of the top of the second support and in its structure is the same as the other portions of the sides of the upper section to be described.

Up to the top of the first cross bar from the top, the upper part of the crate is lined with galvanized iron and from there, up and over the top with wire netting. The lower end of the netting is below the iron. As shown in figure 2 the galvanized iron comes down just over the quarter-sawed piece at the base and in this way any urine that may hit the walls is forced to drop into the central funnel.

In the front of the upper section there is a sliding iron door which is used at feeding time. It runs in grooves at the side and is kept in place by a pin. When the feed box has to be used this sliding door is pulled up and pinned near the top of the crate.

The feeding box is shown in figure 2 and is used only for a short feeding period. It is pushed in and kept in place by a pin in each side of the rear bottom portion.

The other equipment consists of a heavy galvanized iron grating which rests on the upper galvanized ledge of the lower section as shown in figure 2. The mesh, strength and size of wire, and dimensions to be used in this will be determined by the animals used. This bears the weight of the animal and the feces and urine passed through on to a screen of gauze resting on the ledge below. Here the feces are retained and the urine passes through the funnel to the collecting receptacle below. Two pins on each side prevent the upper section from moving on the track except as desired.

Additional numbers of the lower section should be provided. In fact it is better to have two lower sections for each upper. Then when the daily changing of the experimental animals occurs they can be cleaned off and moved on to the new bottoms. The feces, urine and wash water from the bottoms that have been in use are collected and all parts of the lower section thoroughly cleaned and dried. Descriptions for the use of crates of similar types are available so the details regarding the complete collection of the excreta are not required.

BACTERIA COUNTS OF MILK POWDER AS OBTAINED BY THE MICROSCOPIC METHOD¹

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Since recent advances of the powdered milk industry have stimulated the interest of various professional groups to a degree not heretofore attained, it is not unnatural that some attention should be given to the bacterial content of this product, not only from the standpoint of numbers of living organisms present, but also from the standpoint of numbers originally contained in the milk from which it was manufactured. In fact, it has already been suggested (1) that bacterial limits should be set for liquid milk which is to be converted into powder. Irrespective of whether or not control measures are adopted which will specifically designate bacterial limits for liquid milk dried, it will be of interest to know just in how far our present methods of enumerating bacteria are of value for indicating the bacterial content of the liquid material when those methods are applied to the desiccated product. Obviously, the plate method cannot be used for this purpose, therefore the microscopic method as now used, or some modification of it, is the only alternative method which need be considered. The advantages and limitations of this method for the examination of liquid milk have been set forth in numerous publications from the New York Agricultural Experiment Station (Geneva) and need not be enumerated herein.

The adaptability of the microscopic method for the purpose under consideration,—namely, for determining the bacterial content of liquid milk used for drying by the examination of the desiccated product after reconstituting with water,—must be judged in the light of certain limitations in the method itself,

¹ Read before the Society of American Bacteriologists, Philadelphia, December 28, 1921.

and with regard to certain conditions attending the preparation of the product. Notwithstanding the statements occasionally made to the contrary, the microscopic method as now used is not accepted as being reliable for pasteurized milk. Hastings and Davenport (2) have shown that in some instances only 3 per cent of the bacteria found in raw milk can be detected by this method after pasteurization; whereas, in other samples as high as 87 per cent of those originally counted in the raw milk may be found in the pasteurized milk. These wide discrepancies between the countable bacteria before and after pasteurization seem to be due, to some extent, to the species present. From the fact that the microscopic method gives irregular results with heated milk it would seem that any attempt to apply it to reconstituted milk powder would likewise give results of doubtful value for indicating the bacterial content of the liquid milk prior to drying.

Furthermore, it is entirely possible that the degree of heat and the length of time for which the milk is exposed to high temperatures may determine to some extent the relationship between the counts obtained from the raw and heated milk. If such were found to be the case the value of the microscopic method for desiccated milks as a whole would be subject to even further limitations than those imposed by pasteurization, due to the fact that in each of the methods now commonly employed for drying milk, wide variations in temperature and period of exposure are used. Therefore, even if it were possible to state with accuracy that the microscopic method gave a true picture of the bacterial quality of the liquid milk prior to drying by a certain process, or even if it gave constant results as proved by careful comparisons showing a uniform reduction in numbers, it is doubtful whether such conclusions would be valid when the method was applied to samples dried by a different process.

Aside from these probable limitations, there still remains the further handicap of varying solubility of different samples as determined by method of manufacture and by subsequent absorption of moisture from the atmosphere during storage. This factor of insolubility tends to invalidate results by preventing proper performance of the technique rather than by directly affecting the visibility of the organisms present.

EXPERIMENTAL WORK

Even though the microscopic method may appear to have but little value for determining the bacterial content of the original liquid milk when applied to the desiccated product, certain comparisons have been made at this Laboratory for the purpose of ascertaining the actual relationship between the counts obtained from liquid milk and the counts obtained from the same milk after drying by the Just hot-roller process. The particular object of these comparisons was to determine whether this method could be relied upon to indicate gross differences in bacterial content of milk dried each day at different factories, thereby furnishing a basis upon which a control system might be established which would be at least applicable to milk powder made by the Just process.

While there is some doubt as to the inherent value of statements which designate a definite number of bacteria per cubic centimeter, there are certain instances which demand that such expressions be used, and that the numbers recorded be determined as accurately as the particular circumstances may dictate. In making comparisons of bacterial counts from milk by the microscopic method before and after drying, it has been necessary to obtain figures designed to show the actual difference in the number of bacteria found in the respective samples. These comparative counts were obtained in the following manner:

Convenient amounts of the experimental liquid milk (400 to 500 pounds) were placed in an especially devised tank which was attached directly to the feed pipe of a Just process drying machine. After sampling, the milk was turned on and when it had reached the normal level in the trough formed by the revolving cylinders and drying conditions had become normal in other respects, samples of the film of dry milk solids were taken immediately after being cut from the cylinders, this film being subsequently reduced to a fine powder. Soon after the samples were taken duplicate smears from both liquid and dried products were made for microscopic examination. The technique regularly employed for liquid milk was used, the samples of powder having first been diluted with water to give the same concentration of milk solids

as was found in the natural milk. One hundred microscopic fields were examined on each of the duplicate smears by two different analysts. Groups of bacteria consisting of one or more organisms as well as total number of organisms were counted. The results obtained from these examinations are shown in table 1.

From the results given in table 1 it is to be noted that there is a lower number of bacteria in all samples of reconstituted milk than was found in the original liquid milk in all cases except one. Excluding this single sample, the number which could be found in the desiccated milk varied from 30 per cent to 78 per cent of the total found in the original liquid milk; the average from all samples being 45.3 per cent. These results are in fair agreement with those found by Hastings and Davenport (2) from pasteurized milk, and are particularly significant in that they were obtained from a product dried by a process in which the milk is exposed to a temperature above the boiling point for a few seconds.

In reviewing the counts for groups of bacteria consisting of one or more organisms, a remarkably close agreement is found in four of the samples. In two of the samples the group count is higher in the reconstituted milk than in the natural liquid milk. In the remainder of the samples the group count is materially lower in the reconstituted milk. The only interpretation which can be made from these results, is that, there is a tendency for clumps of bacteria to be broken up during the drying process with the consequent result that in the desiccated product there are more organisms existing singly or in small groups of 2, 3, or 4 organisms than were present in the original milk. This difference is readily illustrated by comparing the average size of groups in the two samples. Such a comparison shows that in every sample the average size of the groups is smaller in the desiccated milk than in the natural milk. The average number of organisms per group from all samples of natural milk is 2.7, whereas the average for the desiccated samples is 1.7.

If the microscopic method were to be applied to reconstituted dry milk samples as a routine procedure for determining the general grade of milk dried, or for obtaining information sup-

TABLE 1
Microscopic counts of liquid and dry milk

SAMPLE	ANALYST	SMEAR	NATURAL LIQUID MILK (BACTERIA PER CUBIC CENTIMETER)		RECONSTITUTED DRY MILK (BACTERIA PER CUBIC CENTIMETER)	
			Total count	Group count	Total count	Group count
I	A	1	1,680,000	820,000	1,250,000	500,000
		2	2,750,000	980,000	1,060,000	560,000
	B	1	3,100,000	1,500,000	1,500,000	1,050,000
		2	4,700,000	1,950,000	2,450,000	1,350,000
Average.....			3,057,000	1,312,500	1,565,000	865,000
II	A	1	2,000,000	550,000	770,000	370,000
		2	1,900,000	450,000	1,410,000	510,000
	B	1	3,500,000	1,500,000	2,000,000	1,300,000
		2	5,000,000	1,850,000	2,300,000	2,000,000
Average.....			3,100,000	1,087,000	1,870,000	1,045,000
III	A	1	8,950,000	3,450,000	9,450,000	6,300,000
		2	11,200,000	5,200,000	11,250,000	6,300,000
	B	1	10,250,000	4,100,000	11,650,000	7,100,000
		2	14,900,000	4,950,000	13,500,000	7,600,000
Average.....			11,325,000	4,450,000	11,460,000	6,825,000
IV	A	1	3,610,000	1,400,000	1,560,000	1,130,000
		2	3,840,000	1,330,000	2,270,000	1,170,000
	B	1	4,300,000	1,450,000	2,900,000	1,150,000
		2	4,400,000	1,250,000	3,000,000	1,300,000
Average.....			4,037,500	1,357,000	2,432,500	1,187,500
V	A	1	36,640,000	19,560,000	9,510,000	6,630,000
		2	42,850,000	21,770,000	17,730,000	11,020,000
	B	1	35,000,000	13,800,000	9,700,000	5,400,000
		2	39,600,000	16,500,000	14,350,000	8,550,000
Average.....			38,522,000	17,907,000	12,822,500	7,900,000
VI	A	1	166,600,000	78,500,000	44,600,000	26,990,000
		2	147,100,000	67,600,000	46,730,000	26,820,000
	B	1	154,500,000	52,500,000	56,400,000	31,600,000
		2	160,000,000	46,700,000	52,200,000	26,800,000
Average.....			157,050,000	61,325,000	49,982,500	28,052,500

TABLE 1—Continued

SAMPLE	ANALYST	SMEAR	NATURAL LIQUID MILK (BACTERIA PER CUBIC CENTIMETER)		RECONSTITUTED DRY MILK (BACTERIA PER CUBIC CENTIMETER)	
			Total count	Group count	Total count	Group count
VII	A	1	2, 930, 000	1, 470, 000	1, 600, 000	770, 000
		2	3, 600, 000	1, 730, 000	1, 370, 000	830, 000
	B	1	5, 650, 000	2, 750, 000	1, 100, 000	550, 000
		2	5, 000, 000	1, 950, 000	1, 200, 000	650, 000
Average.....			4, 295, 000	1, 975, 000	1, 317, 500	700, 000
VIII	A	1	3, 130, 000	1, 570, 000	800, 000	570, 000
		2	4, 150, 000	1, 720, 000	1, 600, 000	940, 000
	B	1	3, 850, 000	1, 750, 000	1, 100, 000	750, 000
		2	4, 350, 000	2, 050, 000	2, 000, 000	11, 200, 000
Average.....			3, 870, 000	1, 772, 500	1, 375, 000	865, 000
IX	A	1	17, 700, 000	9, 140, 000	19, 000, 000	11, 750, 000
		2	17, 600, 000	9, 590, 000	9, 500, 000	6, 360, 000
	B	1	14, 350, 000	6, 950, 000	8, 880, 000	5, 500, 000
		2	15, 550, 000	7, 150, 000	13, 650, 000	8, 400, 000
Average.....			16, 300, 000	8, 207, 500	12, 807, 500	8, 002, 500
X	A	1	3, 280, 000	810, 000	3, 150, 000	1, 090, 000
		2	4, 760, 000	1, 120, 000	2, 150, 000	670, 000
	B	1	3, 500, 000	850, 000	1, 950, 000	700, 000
		2	7, 950, 000	1, 200, 000	2, 100, 000	800, 000
Average.....			4, 872, 500	995, 000	2, 337, 000	815, 000
XI	A	1	6, 650, 000	1, 910, 000	3, 300, 000	1, 150, 000
		2	6, 710, 000	2, 160, 000	3, 080, 000	1, 100, 000
	B	1	8, 450, 000	2, 000, 000	3, 950, 000	1, 650, 000
		2	8, 750, 000	2, 450, 000	5, 150, 000	1, 850, 000
Average.....			7, 640, 000	2, 130, 000	3, 870, 000	1, 437, 000
XII	A	1	46, 270, 000	12, 120, 000	35, 460, 000	20, 300, 000
		2	42, 820, 000	11, 120, 000	27, 890, 000	16, 500, 000
	B	1	75, 700, 000	13, 400, 000	51, 100, 000	33, 800, 000
		2	82, 000, 000	15, 200, 000	51, 100, 000	29, 500, 000
Average.....			61, 697, 000	12, 955, 000	41, 387, 000	25, 025, 000
Average all samples.....			26, 311, 000	9, 622, 000	11, 930, 000	6, 892, 000

plementary to bacterial counts regularly determined on milk delivered by patrons, it would be impracticable to spend the time necessary to count the bacteria in 200 microscopic fields as was done with the samples recorded in table 1. In order to ascertain the reliability of counts obtained by the examination of a minimum number of fields, the twelve samples from which 100 fields had been counted on duplicate preparations were examined after an interval of six months without the analyst having knowledge of the results obtained from the detail counts. In the

TABLE 2

Comparisons of results obtained from examination of 200 microscopic fields and 10 microscopic fields

SAMPLE	NATURAL LIQUID MILK (BACTERIA PER CUBIC CENTIMETER —GROUP COUNT)		RECONSTITUTED DRY MILK (BACTERIA PER CUBIC CENTIMETER —GROUP COUNT)	
	200 fields	10 fields	200 fields	10 fields
I	1,312,000	750,000	865,000	650,000
II	1,087,000	1,025,000	1,045,000	900,000
III	4,450,000	4,275,000	6,825,000	4,975,000
IV	1,357,000	1,125,000	1,187,000	1,250,000
V	17,907,000	9,875,000	7,900,000	6,625,000
VI	61,325,000	54,500,000	28,052,000	38,625,000
VII	1,975,000	2,225,000	700,000	1,300,000
VIII	1,772,000	2,350,000	865,000	1,400,000
IX	8,207,000	8,675,000	8,002,000	9,625,000
X	995,000	700,000	815,000	750,000
XI	2,130,000	1,975,000	1,437,000	1,450,000
XII	12,955,000	6,400,000	25,025,000	36,000,000

second examination only groups of bacteria were counted in 5 fields of each of the duplicate preparations. The average results of these examinations compared with those obtained by examining 100 fields from duplicate preparations are shown in table 2. These results would seem to indicate that for the purpose of determining the relative bacterial content of either raw or desiccated milk, there is very little to be gained for control purposes by counting a large number of fields.

In order to secure further information showing the difference in bacterial count of raw liquid milk and reconstituted dry milk

as determined by the practicable method of counting groups in 5 microscopic fields, additional samples were obtained daily for a period of one month and handled in all respects as would be

TABLE 3

Comparisons of number of groups of bacteria in natural and desiccated milk as determined by practicable methods of examination

SAMPLE	NATURAL LIQUID MILK (BACTERIA PER CUBIC CENTIMETER)	RECONSTRUCTED DRY MILK (BACTERIA PER CUBIC CENTIMETER)
1	2,040,000	840,000
2	1,680,000	900,000
3	3,300,000	1,320,000
4	1,800,000	1,080,000
5	5,100,000	1,140,000
6	2,160,000	540,000
7	2,270,000	840,000
8	3,180,000	660,000
9	4,680,000	1,860,000
10	11,400,000	3,420,000
11	8,100,000	5,400,000
12	3,600,000	1,380,000
13	2,400,000	960,000
14	5,460,000	1,920,000
15	3,300,000	960,000
16	2,220,000	840,000
17	2,640,000	1,080,000
18	1,800,000	960,000
19	5,520,000	1,740,000
20	1,980,000	960,000
21	3,000,000	1,800,000
22	7,800,000	3,000,000
23	1,560,000	480,000
24	6,600,000	3,000,000
25	16,000,000	6,600,000
26	4,980,000	1,800,000
27	2,500,000	2,400,000
28	4,800,000	3,120,000
29	15,000,000	5,040,000
30	3,600,000	1,920,000
Average.....	4,649,333	1,932,000

done under routine control conditions. The results of these comparisons are shown in table 3 from which it will be noted that the same general conditions are found as were obtained from

the samples which have already been recorded. These samples, however, contain fewer bacteria than the others and there was an absence of large groups of organisms in the natural milk. This undoubtedly accounts for the fact that none of these samples showed an increase in the group count from the reconstituted milk. The average groups of bacteria counted in the desiccated samples amounted to 41.4 per cent of the number found in the original milk. The close relationship between this figure and that obtained from the samples in which 200 fields were counted tends to strengthen the conclusion that from milk of normal flora, on an average something less than 50 per cent of the total number of bacteria can be detected by the microscope after drying by the Just process.

Variability of microscopic counts from liquid and dried milk

It is not considered that this investigation would be complete without attempting to show the relative accuracy of the microscopic method for natural milk and for reconstituted dry milks in so far as the technique alone is concerned. Since there are certain factors which might cause variations in the counts from the same sample of reconstituted milk, which are not found in natural milk it has seemed advisable to determine the coefficient of variability from samples of liquid milk and samples of milk powder made by different processes in order that the constancy of this factor for each product may be known. This information would also be of value in interpreting the difference obtained between the counts from natural and reconstituted milk, in that, if the same coefficient of variability should be found from both products, the difference in counts such as those already shown in the foregoing tables would be directly attributable to the disintegration of certain organisms or to lack of ability to take the stain rather than to other factors such as, loss of organisms resulting from failure of the desiccated product to adhere to the slide; invisibility due to poor solubility of the powder; lack of uniform thickness of milk solids; or to other interfering factors which might result from desiccation and subsequent reconstitution.

Breed and Stocking (3) have shown that a coefficient of variability as low as 11.7 may be obtained by the microscopic method for group counts made from milk artificially inoculated in order to eliminate the presence of irregular groups containing a large number of organisms. The high degree of accuracy represented by this figure cannot, however, be accepted as applicable to samples of natural milk which may contain single organisms and irregular-sized groups in varying ratios. In order to ascertain the coefficient of variability from milk with natural groups, the group count was obtained on 9 different samples, 10 different preparations being made from each sample. The groups of

TABLE 4
Variations in group counts obtained from normal milk

SAMPLE	AVERAGE BACTERIA PER CUBIC CENTIMETER	STANDARD DEVIATION	PROBABLE ERROR	COEFFICIENT OF VARIABILITY
1	18,000	$\pm 11,400$	7,200	61.2
2	78,000	$\pm 49,800$	33,000	63.8
3	90,000	$\pm 45,000$	30,600	52.8
4	66,000	$\pm 34,000$	22,800	51.8
5	5,880,000	$\pm 3,120,000$	2,100,000	53.0
6	5,220,000	$\pm 2,640,000$	1,740,000	50.5
7	93,000,000	$\pm 14,880,000$	10,000,000	16.0
8	49,800,000	$\pm 13,800,000$	9,300,000	29.0
9	40,200,000	$\pm 16,380,000$	11,040,000	40.7
Average.....				46.3

bacteria were counted in 100 fields on each of the ten smears from each sample. The average count for each sample, the standard deviation, probable error and coefficient of variability are shown in table 4.

Since the samples in table 4 represent wide variations in bacterial quality, it is believed that the average coefficient of variability of 46.3 represents very closely the degree of accuracy which may be expected from the microscopic count of normal samples of liquid milk.

In order that the coefficient of variability from samples of reconstituted milk might be compared with that obtained from milk with natural bacterial groups, samples of powder made by

different processes and with wide variations in bacterial content were selected and the group counts determined. The plan used for obtaining these counts follows:

Each of the samples of dry milk were divided into four or five parts and the necessary amount of water added to each portion to reconstitute to the concentration of the original liquid milk. From each of these separate samples several preparations were made. Not less than 5 microscopic fields were examined from each preparation. The average result from the 5 fields was taken

TABLE 5

Bacteria counts and coefficient of variability obtained from different samples of milk powder

SAMPLE NUMBER	PROCESS OF MANUFACTURE	AVERAGE BACTERIA PER CUBIC CENTIMETER (GROUP COUNT)	COEFFICIENT OF VARIABILITY
1	Hot roller (Just).....	7,320,000	36.0
2	Hot roller (Just).....	11,370,000	23.6
3	Hot roller (Just).....	14,540,000	39.4
4	Hot roller (Just).....	199,900,000	22.0
5	Hot roller (Just).....	42,900,000	13.6
6	Hot roller (Just).....	295,000	68.4
7	Hot roller (Just).....	89,000	104.9
8	Hot roller (Just).....	15,732,000	20.9
9	Kunick modified.....	88,956,000	5.9
10	Kunick modified.....	2,580,000	17.8
11	Kunick modified.....	1,836,000	21.3
12	Spray.....	9,780,000	15.3
13	Spray.....	7,656,000	28.8
14	Spray.....	2,764,000	40.2
Average.....			32.7

as the count for the sub-sample. The results from the different sub-samples were used for determining the coefficient of variability for that particular sample of powder. In table 5 is shown the average bacteria per cubic centimeter (group count) of reconstituted milk and the coefficient of variability as determined from the counts as already explained. From the results shown in this table it is to be noted that the average coefficient of variability from determinations made upon 14 different samples is 32.7. This figure indicates even a greater uniformity of results than is

to be expected from natural liquid milk providing the coefficient of variability of 46.3 from such samples is to be accepted as a criterion of uniformity. It is significant that there is a relatively close agreement in the coefficients of variability from each type of milk powder when the results are over a million bacteria per cubic centimeter.

CONCLUSIONS

The following conclusions may be drawn from the results reported herein:

1. There is a decrease in the number of bacteria which can be detected by the microscopic method in reconstituted milk powder dried by the Just process. The decrease in total numbers as determined by examining 100 fields from duplicate preparations is not constant; variations from 30 per cent to 78 per cent of the number originally present in the natural milk were found in the samples of reconstituted milk. The average number found from all samples of reconstituted milk was 45.3 per cent of those counted in the natural milk.

2. There is a tendency for groups of bacteria existing in natural milk to be broken up during the drying process. Group counts, therefore, may not be reduced in the same proportion as the total count. Relationship between group counts from natural and reconstituted milk powder depends upon the prevalence of large groups in the natural milk. In some instances the group count from reconstituted milk is higher than the group count from natural milk. From samples of natural milk which do not contain excessive numbers of large groups, there was a reduction in group count very similar to the reduction in total count.

3. For practical routine examination of natural or reconstituted milk for control purposes in which it is desired to grade samples into three or four classes, the average count obtained from 10 microscopic fields gives practically as satisfactory results as does the examination of 200 fields.

4. The microscopic method as applied to samples of reconstituted milk gives results which are fully as uniform as those obtained from natural milk when the adaptability of the method

for discerning visible organisms is alone concerned. This statement does not apply to old samples of powder which have become insoluble and thus prevent proper performance of the regular technique.

5. Microscopic examinations of milk powder made by the hot roller process are justified as a partial control measure for commercial laboratories in which it is desired to ascertain the relative bacterial content of the milk dried. The results however, must be interpreted with due recognition of the fact that a certain percentage have disappeared during the process; the results reported herein show that the average decrease has amounted to approximately 50 per cent.

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